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MORPHOLOGICAL AND ECOLOGICAL STUDIES ON HEMIPTERA: COCCOIDEA

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RIASSUNTO

Le cocciniglie (Hemiptera: Coccoidea) costituiscono una Superfamiglia di fitomizi di interesse economico, che attaccano piante coltivate di intereresse agrario e forestale, sia in condizioni di pieno campo sia in serra. Tale gruppo di insetti colonizza gli habitat in tutte le regioni del mondo e a tutte le latitudini, tranne che nelle regioni polari. Sebbene rivestano un ruolo chiave nell'ecologia dei sistemi agrari e di foresta, la Superfamiglia Coccoidea rappresenta un gruppo tassonomico poco studiato. La presente dissertazione approfondisce due temi di ricerca affrontati nel corso del dottorato, e precisamente la morfologia (capitolo I) e studi di bio-ecologia e biologia (capitolo II). Il lavoro svolto nei due diversi ambiti fornisce strumenti originali per l'identificazione di specie e informazioni inedite sulla biologia di due cocciniglie. I taxa oggetto di studio sono afferenti alle famiglie Coccidae e Pseudococcidae. Per quanto concerne gli aspetti tassonomici la ricerca ha affrontato lo studio della morfologia degli stadi maschili e degli stadi giovanili, i quali recentemente sono stati oggetti di particolare interesse per la loro utilità nel campo sistematico. Attualmente, infatti, la sistematica della superfamiglia Coccoidea si basa quasi esclusivamente sui caratteri morfologici della femmina adulta.

I primi due lavori di morfologia riguardano due Coccidi Ceroplastini, afferenti rispettivamente alle specie *Ceroplastes japonicus* Green (sezione I) e *Ceroplastes rusci* L. (sezione II). Nei suddetti lavori si descrive la morfologia dei maschi adulti e degli stadi neanidali maschili. Viene infine fornita una chiave di riconoscimento basata sui caratteri dei pochi maschi adulti di Ceroplastini ad oggi descritti e sono quindi discusse le affinità tra le specie.

Un ultimo lavoro di tassonomia riguarda lo studio della morfologia del Coccide Cardiococcino messicano *Ceroplastodes dugesii* (Signoret), di cui si sono descritti tutti gli stadi di sviluppo, maschili e femminili; ad esclusione del maschio adulto. Il suddetto lavoro fornisce nuovi strumenti tassonomici per l'identificazione della specie; in particolare la descrizione della morfologia della femmina è integrata riportando caratteri, mai osservati in precedenza. La presenza di bande divergenti di pori preopercolari presenti sul dorso, evidenziati in questo studio, assume quindi valore diagnostico.

Nel capitolo II sono riportati due studi sull'ecologia di due specie ritenute dannose alle piante ornamentali e ai fruttiferi. Nella sezione I viene trattato Parthenolecanium rufulum (Cockerell) (Coccidae), mentre lo studio di Pseudococcus comstocki (Kuwana) (Pseudococcidae) è trattato nella sezione II. Le indagini su Parthenolecanium rufulum affrontano la fenologia e la morfologia di questa occiniglia, specie a diffusione paleartica infeudata prevalentemente alle querce a foglia caduca. La specie è stata monitorata nel periodo luglio 2006 – 2008 su piante di Quercus robur. P rufulum è specie monovoltina con svernamento allo stadio di neanide di 2^a età sui rami. La muta ad adulto avviene a partire da metà aprile. L'ovideposizione avviene tra la terza decade di aprile e la seconda decade di maggio. La schiusura delle uova si verifica sin dalla fine di maggio e le neanidi di 1^ª età (crawlers) migrano dai rami verso la pagina inferiore delle foglie. Nel corso del mese di agosto avviene la muta a neanide di 2^ª età. A partire da metà settembre le neanidi di 2^ª età migrano gradualmente dalle foglie ai rami, ove svernano. La migrazione si completa durante la prima decade di dicembre. Dai risultati del presente studio si evince che la fenologia della cocciniglia appare anticipata di circa un mese rispetto a quanto noto per il centro Europa. Infine sono riportate alcune note sulla presenza di Anthribus nebulosus Forster (Coleoptera: Anthribidae), predatore di P. rufulum. Oltre allo studio della fenologia di P. rufulum, è riportata la descrizione morfologica degli stadi neanidali, finora descritti solo in modo incompleto.

Nella sezione II è riportato lo studio su *Pseudococcus comstocki* (Kuwana), specie esotica di recente introduzione in Italia, infeudata a numerose specie di fruttiferi e ornamentali. I primi risultati di un monitoraggio sulla presenza di *P. comstocki* nel territorio Veneto è stata condotta nell'estate 2008. I risultati sono riportati nella sottosezione I. Sono stati individuati numerosi focolai di infestazione su ornamentali e in particolare su piante di gelso (*Morus nigra*); sono stati rinvenuti inoltre nuovi focolai nell'ambito dei pescheti. I risultati dell'indagine suggeriscono un'ampia diffusione dello pseudococcide sul territorio.

Ecologia e biologia dello pseudococcide sono state studiate sia in pieno campo sia in condizioni controllate di screen-house e laboratorio. Le osservazioni condotte in

screen-house sulla fenologia di P. comstocki (2007-2008) confermano lo svernamento allo stadio di uovo e il verificarsi di 3 generazioni all'anno. La nascita delle neanidi di prima età (crawlers) avviene rispettivamente in aprile, giugno e ottobre. Oltre ai dati ottenuti da popolazioni allevate in screen-house, colonie di P. comstocki sono state allevate su patata in celle climatiche a due livelli termici (T_1 = 25 °C e T_2 =30 °). E' stato quindi valutato l'effetto delle due temperature sui parametri delle tavole di sopravvivenza (R_0 , Rm, λ , DT, T) e sulla fecondità e sulla sex ratio. Allo scopo di fornire informazioni utili alla gestione delle infestazioni è stata verificata l'eventuale presenza di nemici naturali (sottosezione III) e l'effetto di alcuni principi attivi in pieno campo (sottosezione IV). Per quanto riguarda l'indagine sui nemici naturali, è stata accertata la presenza di alcuni interessanti imenotteri parassitoidi. Di particolare rilevanza appare la presenza di *Clausenia purpurea* Ishi, (Hymenopera: Encyrtidae), specie finora mai riscontrata in Europa, sfarfallata da materiale raccolto su pesco. Inoltre si segnala il rinvenimento di Chrysoplatycerus splendens (Howard), (Hymenoptera: Encyrtidae), anch'esso parassitoide di P. comstocki. Nella sottosezione IV si riportano i risultati di una prova di lotta chimica su pesco nei confronti di P. comstocki, condotta utilizzando quattro diverse sostanze attive (Thiametoxam, Clorpirifos-etil, Fosmet e Piretro). Solo la tesi trattata con Thiametoxam ha indotto una riduzione significativa dell' infestazione rispetto al gruppo di controllo.

ABSTRACT

Scale insects (Hempitera: Coccoidea) are noxious sap-sucking plant pests, most important as agricultural pest of perennial plants, woody ornamentals, greenhouse plants and forest trees. They are worldwide distributed and occur in almost all habitats. Unlike their ecological and economic importance, scale insects represent a neglected research topic and most studies are almost always referred to the main families, bringing to the current dearth of scientific data for this taxon.

This thesis deals with two main investigation fields: taxonomy (Chapter I) and bioecology (Chapter II) of scale insects. With regard to taxonomy, the work focuses on the adult males and immatures morphology, that may play a critical role in understanding scale insects systematic relationships, currently based on teneral adult females morphology. The information gathered from these studies will be important in the pests management by providing new identification means The first two taxonomical studies concern the morphology of adult males and male instars of *Ceroplastes japonicus* Green and *Ceroplastes rusci* L (Hemiptera: Coccidae: Ceroplastinae). These instars are here described and illustrated for the first time (Section I and II). An identification key to the adult males of six wax scale species is also provided and morphological affinities are here discussed.

Finally, with the recent collection of fresh specimen of the Mexican wax scale *Ceroplastodes dugesii* (Signoret) (Hemiptera: Coccidae: Cardiococcinae), the opportunity is taken to properly redescribe the adult female and the 1st-instar nymph and to describe the remaining male and female stages apart from the adult male. Descriptions and illustrations, reported on section III, complete the adult female morphology, adding previously unreported morphological features, e.g. one pair of distinctive bands of preopercular pores occurring on dorsum.

Ecological studies (Chapter II) regard two species: *P. rufulum* (Cockerell) (Coccidae) (Section I) and *P. comstocki* (Kuwana) (Pseudococcidae) (Section II), both reported as a pests of orchards and ornamentals.

Section I deals with observations on the life history, phenology and morphology of *P*. *rufulum*, a Palaearctic species largely distributed in European countries and especially

common on *Quercus*. The study was carried out in North-eastern Italy on *Quercus* robur trees from July 2006 to June 2008. *P. rufulum* develops one generation/ year and overwinters as 2^{nd} instar nymph. Moulting to adult female occurs from mid April onward. Egg-laying starts between the first days of April to mid May. Egg hatching occurs from the end of May. First-instar nymphs settle on the undersurface of leaves. Moults to 2^{nd} - instar nymph occur in August. Starting from mid September, the 2^{nd} -instar nymphs gradually migrate from the leaves to the twigs to overwinter. This migration is completed by the beginning of December. Phenology pattern in Italy appears earlier than in Central Europe by about one month. The nymphal instars of *P. rufulum* are redescribed and illustrated. Observations on the presence of the predator *Anthribus nebulosus* Forster (Coleoptera: Anthribidae) are reported as well.

Section II deals with *Pseudococcus comstocki*, an Asiatic mealybug, pest of fruit and ornamentals, recently recorded in Italy. A comprehensive study on the bio-ecology of P. comstocki has been carried out under both field and laboratory conditions. Fieldwork has been carried out by a preliminary survey on the occurrence of the Comstock mealybug within the Verona district (North–eastern Italy) during summer 2008 (Subsection I). Many foci of P. comstocki were found on mulberry (Morus nigra) trees. Moreover two infested peach orchards were recorded near Verona, suggesting a widespread distribution of the mealybug. In addition to fieldwork, some aspects of the biology were investigated and life history surveyed on captive colonies of P. comstocki reared under both screen-house and laboratory conditions. Results provided by the survey of *P*. comstocki under screen-house conditions (2007-2008) confirm that it overwinters at the egg stage and develops 3 generations/year. First instar nymphs (crawlers) occur in April, between June and July, and in October. These biological observations gather with data obtained from laboratory. The effect of two temperature regimes (T₁ = 25 °C; T₂ = 30 °C) on life table parameters (R_0 , Rm, λ , DT, T) fecundity and sex ratio was assessed. Temperature has significant effect (a < 0.01) on R_0 , Rm, λ , fecundity and sex ratio. The differences were not significant for Dt and T. This study on P. comstocki provides also guidelines for pest management with both biological and chemical methods. A survey of natural enemies of this pest was initiated and a first list of parasitoids of P comstocki is provided (subsection III). The encyrtid wasp Clausenia purpurea Ishii, (Hymenoptera: Encyrtidae) has been

recorded. *C. purpurea*, first record for Europe, is considered as an effective biological control agent of *P. comstocki* and was introduced into Israel and California (USA) from Japan during 1940's. In addition, *Chrysoplatycerus splendens* (Howard), (Hymenoptera: Encyrtidae), native from the Afrotropical Region, was obtained from *P. comstocki* infesting *Viburnum tinus* hedges and currently represent the first record of this species in the Palearctic Region. Finally, the effect of Thiametoxam, Chlorpyriphos-ethil, Phosmet and Pyrethrum was assayed on *P. comstocki* in a preliminary trial in one infested peach orchard (subsection IV). Satisfactory results were obtained for only Thiametoxam.

GENERAL INTRODUCTION

Scale insects (Hemiptera: Sternorrhyncha: Coccoidea) are an important taxon of about 7.000 species grouped into 20 to 30 families, depending to the authority (Hodgson & Foldi, 2006; Miller & Davidson, 2007). They are obligate sap-sucking and generally sedentary plant-parasites occurring worldwide in nearly all habitats, from the tundra to the tropics, except polar areas (Ben Dov, 1997). Scale insects are related to Aphidoidea (aphids), Psylloidea (jumping plant lice), Aleyrodidoidea (whiteflies). There is currently a considerable debate among scale workers about the appropriate level of classification of the scale insects; the question is whether they should be a superfamily (Coccoidea) or a separated suborder (Coccinea), due to their unusual and almost unique features (Hodgson& Foldi, 2006; Lambdin, 2001; Morrison, 1928;).

Scale insects have a very distinct sexual dimorphism: adult females are wingless, saclike, ovoid or circular in shape, present unclear separation between head, thorax and abdomen, and they may or may not have legs. Male scale insects are midge-like and usually have one pair of wings with reduced venation. The second pair is reduced to hamulohalteres. Adult females emerge directly from the 2nd- or 3rd-instar nymphs (according to species), whereas males have four nymph instars and undergo a metamorphosis of the neometabola type (Ben Dov, 1997): from nymph instars through prepupa (or third instar) and pupa (or fourth instar) to adult.. Most scale insects produce waxy secretions that accumulate on the body surface, appearing like a thin translucent sheet or a thick wet mass or a powdery wax. Finally armoured scale insects are easily recognised in the field by producing a unique protective scale cover, that is detached from the body.

Scale insects are noxious plant pests, important as agricultural pest of perennial plants, woody ornamentals, greenhouse plants and forest trees (Kosztarab, 1996; Ben Dov, 1997). Damage is usually produced by removal of plant sap or direct damages to tissues, but also may be caused by transmission of plant pathogens (Conti, 2004; Sforza, 2003; Pellizzari, 1997) and by the excretion of large quantities of honeydew with subsequent growth of sooty mould fungi that cover leaf surfaces and reduce photosynthesis (Kosztarab, 1996; Ben Dov, 1997). Scales also can be beneficial, since they have been a significant economic income source. Especially scale insects red dyes

were historically very important for man activities: in the Mediterranean area, kermesids (Hemiptera: Kermesidae) growing on oaks (particularly on Quercus coccifera), have been used in Europe and Asia since Roman times or before, when the Phoenician shellfish purple dye industry declined (Ferreira et al, 2004). Kermesids were replaced by the introduction to Europe by the Spaniards, in the 16th century, of the American Cochineal, obtained from the cochineal scales (Hemiptera: Dactylopyiidae), though it had been used in South and Central America long before (Ferreira et al, 2004). Despite this common deploy since the antiquities, scale insects were not recognised as insects for a log time and commonly mistaken for plant organs or galls: the same Latin name for scale insects derives from the Greek kokkos, translated in Latin as *coccus* or *granum*, that means seed or berry, due to their shape. Despite their ecological and economic importance, scale insects represent a neglected research topic. Bio-ecological and systematics studies are scarce and almost referred to the main taxa. The current dearth of scientific data is mainly due to studying difficulties since their small dimensions and highly cryptic habits. For these reasons they are difficult to detect, especially at low population densities and subsequent species identification is frequently inappropriate due to the absence of solid taxonomy background.

CHAPTER I

MORPHOLOGICAL STUDIES

INTRODUCTION

Use of adult males as a taxonomical tool

Although a vast amount of work has already been done on the description and taxonomy of female scale insects, the study of adult males and immatures is still largely neglected. There are various reasons for the dearth of information on the structure of male scales. Adult males are generally underused because they are small, fragile, and infrequently collected since their life-span is brief and appears seasonally, with very short emergence time. Furthermore, in some species males are absent or rarely occurring. Adult males morphology may play a critical role in understanding scale insect systematic relationships, currently based on adult females morphology. Coccoidea adult females are usually neotenic, after they retain a preimaginal appearance and adulthood is reached without deep morphological change; some scale insect families, such as Diaspididae, are catametabolic and adults undergo through a morphological simplification during post-embrionic development. In both cases there are few characters that can be used to consider the possible relationships between taxa. Unlike neotenic or catametabolic females, adult males are fully mature insects especially appropriate for systematics analysis (Miller & Kosztarab, 1979), as the homology of most of the characters is reasonably clear (Hodgson & Foldi, 2006). This special field of study was took into account also by Ferris (1937) who considered adult males characters of some aid in general classification.

Unfortunately such taxonomical approach is difficult because of the lack of works based on adult males morphology: the adult males of perhaps only about 60 species have been described adequately up to 2004 (Hodgson, 2004), but, more recently, many significant steps forwards were made, especially by Hodgson & Foldi (2006), Hodgson (2005), Hodgson & Henderson (2004). Furthermore, most of the available information on male adults is found in early papers containing brief notes on more obvious features of their anatomy and seldom contain important morphological details. For this reason it may be difficult to analyse relationships between taxa. Finally, males identification is often doubtful, because more than one species may share the same host plant and male crawlers are known to settle sometimes some distance from where the female crawlers settle, even on different parts of the same plant (Hodgson& Foldi,

2006). For this reason the species identity of each collection should be confirmed from their association with adult females and care should be taken with the choice of specimen in the case mixed collections.

In the present study adult males descriptions are provided for *Ceroplastes japonicus* Green and *Ceroplastes rusci* L. (chapter I, sections I & II).

Other taxonomical tools

Many current interests lie in systematics of the Coccoidea based on taxonomic characters alternative to those of teneral adult females. In some cases tests produced by males during their development can also provide an alternative means of identification (Miller and Williams, 1997). The second instar male secretes a protective wax cover, called male test, under which the prepupa and pupa stages develop and protect themselves from environmental stress such as dehydration or natural enemies. Each scale family of scale insect is characterised by a different type of male covering, e.g. a scale cover in armoured scale insects (Diaspidae), a fluffy cocoon in mealybugs (Pseudococcidae) or a glassy wax cover in soft scales (Coccidae). Unfortunately only the soft scales present morphological features of some diagnostic value at specific level. Some authors used coccids male tests for species identification (Rehacek, 1960; Richard, 1958) and, more recently, significant steps in this direction were made by Miller and Williams (1990) and Henderson and Rhode (2001). Many interests also lie in systematics based on immature description for diagnostic purposes. Identification keys based on immatures stages are needed for practical reasons, most due to the fact that young adults may occur for only a few days each year, hence scale insects occurring on host plants are often nymphs. Furthermore, the correct timing of collection is essential for obtaining suitable material, since older females are often too heavily sclerotised for study. Gullan (2000) produced an identification key to six mealubugs occuring on citrus in Australia, based on immatures characters, Miller (1991) studied affinities within sixteen families according to 1st-instar characters, and Howell and Tippins (1990) investigated taxonomical relationships between immature armoured scales. This research field mainly concerns the first instar nymphs, but only a little work is done about the other immature stages (Williams and Hodge, 1997). Furthermore, only small groups of the

Coccoidea, mainly coccids and armoured scales, have some complete nymph instars descriptions.

With regard to the present work, a small contribution to alternative taxonomy analysis is provided. Descriptions of nymph instars are provided for *Ceroplastes japonicus* Green, *Ceroplastes rusci* L., *Ceroplastodes dugesii* (Signoret, 1893) (chapter I, sections I & II), *Parthenolecanium rufulum* (Cockerell) (chapter II, section I). The male test of *Ceroplastes japonicus* Green has been described as well (chapter I, sections I).

Material and methods

All specimens were slide mounted according to the procedures of Ben-Dov and Hodgson (1997). Measurements of specimens were made using an ocular micrometer (Meopta H8x M10) on a Jenamed phase-contrast microscope. Measurements and numbers are given as ranges, followed by the mean in parentheses. Terminology follows that of Giliomee (1967). With regard to the *Ceroplastes japonicus* male test, terminology follows that of Miller and Williams (1990). The illustrations of the coccids follow the conventional style used for the Coccoidea, with the dorsal surface of the body depicted on the left side of the drawing and the ventral surface on the right. The vignettes around the margin show some important features enlarged, and these are not drawn on the same scale. Some structures, such as phragma and other internal structures, lye deeply within the body; it is the convention to show these using dashed lines or with schematic draws with complete lines.

Specimen depositories: mounted and unmounted specimens are deposited in the entomological collection in the Department of Environmental Agronomy and Crop Production–Entomology (DEAE), University of Padua, Italy.

SECTION I

REDESCRIPTION OF THE ADULT MALE AND DESCRIPTION OF SECOND-INSTAR MALE, PREPUPA AND PUPA OF *CEROPLASTES JAPONICUS* GREEN

(HEMIPTERA: COCCOIDEA: COCCIDAE)

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Introduction

The genus *Ceroplastes* comprises many widespread and economically important wax scale species, mainly pests of tropical and subtropical fruit trees, citrus, fig, ornamentals (e.g. C. sinensis Del Guercio, C. rusci (Linnaeus), C. rubens Maskell, C. destructor Newstead, C. floridensis Comstock). This genus includes both bisexual species, where males are known (i.e. C. sinensis, C. rusci), even if only rarely recorded (i.e. C. cer- iferus (Fabricius) (Gimpel et al. 1974), and species that can reproduce parthenogenetically. Parthenogenetic reproduction may be either facultative, where males only occur rarely, or obligatory, where males do not occur, as with C. floridensis and C. destructor (Qin & Gullan, 1994). Differences in the presence or absence of males in populations of a given species living in different geographic areas have also been reported. For instance, according to Kuwana (1923), males of C. rubens are present in Japan, but they have not been recorded in the USA (Gimpel et al., 1974), nor in Australia (Qin & Gullan, 1994). With regard to Ceroplastes japonicus Green, males are known in the supposed native area of the species (China, Korea, Japan) (Kuwana, 1923; Jiang & Gu, 1988; Park et al., 1992; Xie et al., 2006), and also in Georgia and Russia (Abkhazia), where it is an introduced species (Borchsenius, 1957; Japoshvili, 2001, pers. com.). In Europe, C. japonicus was first recorded outdoors in Italy (Kozár et al., 1984), and later in France, Slovenia and Croatia (Pellizzari & Camporese, 1994; Janar et al., 1999; Masten Milek et al., 2007) In the above recorded countries it is a pest of ornamentals (e.g. Hedera helix, Ilex aquifolium, Laurus nobilis and Citrus) in urban environments. Males of C. japonicus were not noticed in Italy for a long time, even though its biology was studied in different areas by several authors (Longo, 1985; Camporese, 1991; Camporese & Pellizzari, 1998; Raspi & Antonelli, 1998). Although populations of this species have been regularly monitored in Padua (Italy) since 1990, males were not observed until 2003, when male tests were noticed for the first time on Citrus reticulata. At this time (the end of September), several male tests were empty or had dead specimens inside. Subsequently, many adult males, 2nd-instar male nymphs, prepupae and pupae were observed on 14th October 2007 on Laurus nobilis, in the province of Venice. The morphology of the adult male of C. japonicus was at first studied by Borchsenius (1957).

PLATE 1. (a) Star-shaped male test of *C. japonicus* (Green); the test exhibits 13 noticeable lateral waxy projections; (b) Male tests on lower surface of a leaf; (c) Comparison between young females (pinkish, in the middle) and male tests (white); (d) Adult male inside the test, the arrow indicates the peripheral fringe around the test base; (e & f) Adult male emerging backwards from waxy test.



He gave a detailed description and illustration of the head, the third and last antennal segments, scutum and penial sheath. Probably because the description is in Russian and some illustrations are in the first part of his book (p. 22: male; p. 26: head; p. 30: penial sheath; p. 32: male test) and not in the pages dealing with *C. japonicus* (p. 461–468), they were overlooked for a long time. Another description and illustration of *C. japonicus* male was published by Xie *et al.* (2006), but the small size of the illustration and the short description (in Chinese) does not allow a clear understanding of some morphological characters. No descriptions are available of the 2nd-instar male nymph, the prepupa and pupa. With the recent collections, the opportunity is here taken to illustrate and describe the 2nd-instar male, prepupa and pupa, and to give a detailed redescription of the adult male of this species.

Ceroplastes japonicus Green

Ceroplastes floridensis japonicus Green, 1921: 258. Ceroplastes japonicus Green; Borchsenius 1949: 181.

Material examined. ITALY: Padua, on *Citrus reticulata*, ix.2003, G. Pellizzari: 11 adult males, 10 2nd- instar males, 10 prepupae, 11 pupae, 6 male tests; and Noale (Venice), on *Laurus nobilis*, 14.x.2007, A. Rainato: 17 adult males, 10 2nd-instar males, 10 prepupae, 10 pupae + 10 male tests.

MALE TEST (Plate 1: a-d)

Described from 10 male tests; total length and total width measured from 5 specimens in fairly good condition, with undamaged waxy projections; details checked on remaining specimens.

General appearance of male test: white, opaque, with a dry wax structure, oblong, star-shaped; median plate strongly elevated, slightly broadened and more elevated anteriorly; with 13 distinct marginal waxy projections plus 2 small anal plate projections; parastigmal processes not seen on examined specimens. Total length, including waxy projections, 1758–2012 (1874) µm; width 1043–1639 (1416) µm. Test

base well defined, ellipsoidal, with peripheral waxy fringe along margin; length of test base: $835-1363 (1027) \mu m$; width: $461-725 (585) \mu m$.

Distribution on host plant: male tests are usually located on the lower surface of leaves.

Comments. Borchsenius (1957) provided a short description (p. 465) and a drawing (p. 32) of the test of *C. japonicus*. Our description and measurements agree with his (about 2.1 mm in length and 1.6 mm in width).

ADULT MALE (Fig. 1, Plate 1: e, f)

Described from 8 males in fairly good condition; details checked on remaining specimens.

Mounted material: 983–1267 (1132) μ m long; 326–359 (338) μ m wide across triangular plates; body broad and stocky, but this possibly due to specimen becoming squashed when mounted. Body and appendages covered with numerous setae, mostly fleshy setae (*fs*); hair-like setae (*hs*) fewer. Dermal pores entirely absent. Abdominal glandular pouches and associated setae absent. Caudal extensions present on abdominal segments VII and VIII.

Head: Bluntly triangular in dorsal view; length from apex to pronotal ridge (*prnr*) 137–204 (170) μ m, width across genae 179–241 (209) μ m. Median crest (*mc*) showing polygonal reticulations; dorsal head setae with 12–14 (13) *fs* and 4–8 (7) *hs*. Dorsal mid-cranial ridge absent; ventral midcranial ridge (*vmcr*) strong, extending from short lateral arms posteriorly to margin of ocular sclerite; with 4 ventral midcranial ridge *hs*. Genae (*g*) sclerotised, with polygonal reticulations and with 22–27 (24) *fs* and 2–5 *hs* genal setae (*gs*) on each side. Simple eyes round, subequal in size; dorsal eyes (*dse*) situated near head apex, each 24–33 (29) μ m wide; ventral eyes (*vse*) on posterior ventral area of head, each 22–37 (27) μ m wide. Ocelli (*o*) situated laterally, each 14.8–18.5 (15.4) μ m wide, lying just anterior to postocular ridge (*pocr*); interocular ridge absent. Ocular sclerite fairly well sclerotised, with polygonal reticulations throughout.

Preocular ridge quite distinct but short; postocular ridge (*pocr*) strongly developed. Dorsal ocular setae (*dos*) 3 *fs* and 1 *hs*. Ventral head setae 12–16 (14) *fs*, mainly situated antero-laterally to ventral simple eyes. Preoral ridge in some specimens fairly well developed, about 15 μ m in length. Cranial apophysis poorly defined, with bifurcate apex.

Antennae: ten-segmented and filiform, with short, stout fleshy setae (fs); each antenna 372–522 (434)µmlong; shorter than half body length (ratio of total body length to antennal length 1: 0.38–0.41, average 0.38), shorter than posterior leg (ratio of posterior leg length to antennal length 1:0.30) and longer than penial sheath (ratio of penial sheath to antennal length 1:1.52–1.68, average 1.63). Scape (*scp*) approximately square, 26–37 (31) μ m long, 22–56 (37) μ m wide; with 2 or 3 hs, plus sometimes 1 fs. Pedicel (*pdc*), 26–37 (31 µm) long, 33–41 (37) µm wide; with 1–5 (3.5) *fs* and 0–4 (2) hs, and with polygonal reticulations on distal 1/3. Segment III club-shaped, 1.77-2.25(1.79) times longer than wide; 33–59 (39) µm long, 15–33 (22) µm maximum width, with 3–10 (6) fs and 0–2 hs. Segments IV–IX cylindrical, each about 15–28 (21 μ m) wide; lengths (µm): III 15–33 (22); IV 85–111 (97); V 59–85 (71); VI 36–57 (51); VII 15–33 (26); VIII 35–48 (41); IX 33–43 (38); setal distribution: III 3–10 (6), IV 18–28 (23), V 12-18 (15), VI 10-21 (15), VII 5-9 (7), VIII 8-17 (11), IX 6-11 (9) all fs except for 1 or 2 hs on each of segments III, VIII and IX. One antennal bristle (abr) present on segments VIII & IX, each 37-48 (44)µmlong and distinctly larger than fs. Segment X 37–52 (44)µmlong, 15–22 (20) µm wide, with apex constricted; with 2–6 (4) fs, 0-4 hs, 3 abr and 3 or 4 subapical capitatae setae:.

Thorax

Prothorax: membranous; pronotal ridges (*prnr*) well developed, medially fused by weak sclerotisation; pronotal sclerite (*prn*) present; without lateral pronotal setae. Median pronotal setae absent; post-tergite not detected. Prosternum (*stn 1*) lightly sclerotised; with base of median ridge and transverse ridge strongly developed; with total of 8-10 *fs* prosternal setae (*snt 1s*). Anteprosternal setae absent. Antemesospiracular setae: 3 on each side of body.

Mesothorax: mesoprephragma with deep ventral emargination. Prescutum (prsc) 78-111 (94) µm long; 126–161 (139) µm wide; strongly sclerotised, with polygonal reticulations; anterior margin curved; laterally bounded by heavily sclerotised prescutal ridges (pscr) and posteriorly by prescutal suture (pscs). Scutum (sct): with median membranous area trapezoidal or sub-rectangular, wider posteriorly; 26–78 (49) µm long; 148–185 (166) µm wide; scutal setae (scts): 10–14 (12) fs and 6–10 (8) hs. Rest of the scutum sclerotised, with fairly well-defined polygonal nodulation, but without setae. Scutellum (scl) 33-48 (41) µm long, 133-178 (155) µm wide; tubular, with small ventral foramen; without scutellar ridge or scutellar setae. Prealare (pra) and triangular plate (tp) well developed. Tegula (teg) present, with 1-4 (3) hs tegular setae (tegs). Mesopostnotum (pn 2) well developed; postnotal apophysis (pna) and postalare well developed and strongly sclerotised. Postalare (pa) without nodulation and without postalare setae. Mesopostphragma with deep emargination. Mesopleural apophysis and mesopleural wing process well developed. Basalare well developed. Subalare present. Mesepisternum (eps 2) with nodulations; subepisternal ridge well developed. Mesepimeron not seen or not developed. Lateropleurite broad, partly bounded anteriorly by an extension from marginal ridge (mr). Basisternum (stn 2)111–155 (137) µm long, 148–248 (218) µm wide; with a strong medial ridge and bounded anteriorly by strong marginal ridges and posteriorly by strong precoxal ridges (pcr 2); without setae; furca (f) well developed, narrow waisted, with arms divergent and extending about 3/4 way to marginal ridge anteriorly. Mesothoracic spiracle (sp 2) with well-developed peritreme; width of peritreme 13-22 (17) µm; postmesospiracular setae (pms 2): 25-35 (27) fs, arranged in a band across segment between spiracles.

Metathorax: suspensorial sclerite absent. Metapostnotum not detected; with a single *fs* metatergal seta on each side; dorsospiracular setae (*dss*) 4–6 *fs*. Metapleural ridge and metapleural ridge wing process well developed. Metasternum (*stn 3*) quite strongly sclerotised; anterior metasternal setae (*amss*) 12–24 (17) *fs*; posterior metasternal setae (*pmss*) 14–19 (17) *fs*. Metepisternum sclerotised, with 6–8 *fs* postmetaspiracular setae; precoxal ridge well developed, with a short, fairly well sclerotised metepimeron produced posteriorly. Metathoracic spiracle similar to mesothoracic one; width of
peritreme 15-30 (18) µm. Wings: hyaline; rather short and comparatively broad; 864-983 (905) µm long and 373–522 (443) µm wide; ratio of width to length 1:1.89–2.32 (2.04); ratio of total body length to wing length: 0.78-0.88 (0.80); alar lobe and alar setae absent. Hamulohalterae absent. Legs: long and slender, prothoracic leg shortest, metathoracic leg longest; total lengths (µm) : I 844–1027 (946); II 844–1065 (969); III 885–1201 (1089) μm; ratio of hind leg to total body length 1:2–2.2 (1:2.1). Coxae (*cx*): I 44–85 (53); II 59–89 (72); III 59–63 (79)µmlong; setae of coxa III 8–19 (15) fs and 2–9 (6) hs. Trochanter (tr) + femur (fm) lengths (µm): I 174–215 (197); II 167–200 (185); III 174–218 (194); maximum widths: I 26–44 (35); II 31–41 (37); III 33–44 (39); ratio of maximum width to length of hind femur 1:4.9-5.2 (5); with 2 campaniform sensilla on each side of trochanter. Trochanter III with 7-12 (9) fs and 1-3 hs; femur III with 14–31 (22) fs and 2–8 (5) hs. Tibia (ti): I 196–251 (228); II 192– 263 (230); III 233–315 (284)µmlong; tibia III with 42–75 (58) fs; 4–15 (9) hs; with one tibial spur (tibs). Tarsi (tar) one-segmented: I 92-111 (101); II 96-111 (103); III 74–122 (107) µm long (ratio of length of tibia III to length of tarsus III 1:0.31–0.39 (0.38)); tarsus III with 12–43 (26) fs, 2–4 hs; tarsal campaniform pores absent; tarsal digitules (tdgt) each 20– 22 μ m long, with apical knob. Claws (c) short, slightly curved, denticle small or absent : length III 15-22 (19) m, claw digitules (cdgt) capitate, each 20–26 (23) µm long.

FIGURE 1. Ceroplastes japonicus Green, adult male.



Where, A = last antennal segments; B = body setae: fleshy seta (B I), hair seta (BII); C =metatarsus and claw; D = dorsal and ventral view of the genital segment; E = cranialapophysis; F = dorsal (FI) and ventral (FII) view of polygonal reticulations on head capsule. And where abr = antennal bristles; ads = dorsal abdominal setae; aed = aedeagus; ams 3s =antemetaspiracular setae; amss = anterior metasternal setae; asII- asVIII= abdominal sternites II-VIII; atI – atVIII= abdominal tergite I-VIII; bra = basal rod of aedeagus; c = claw; cb = coxal bristles; ccx = costal complex of wing veins; cdgt = claw digitules; ce VII = caudal extension of segment VII; ceVIII = caudal extension of segment VIII; cx 3 = coxa of methathoracic leg; dos = dorsal ocular setae; dpls = dorsopleural setae; dse = dorsal simpleeye; dss = dorsal spiracular setae; eps2 = mesepisternum; f = furca; fm3 = femur of metathoracic leg; g = genae; gfs = genital fleshy setae; gs = genal setae; mc = median crest; mr = marginal ridge; o = ocellus; pa = postalare; pdc = pedicel; pepcv = proepisternum +cervical sclerite; pms2 = postmesospiracular setae; pmss = posterior metasternal setae; pn2 =mesopostnotum; pna = postnatal apophysis; pocr = preocular ridge; pra = prealare; prn = lateral pronotal sclerite; prnr = pronotal ridge; prsc = prescutum; ps = penial sheath; pscr = prescutal ridge; pscs = prescutal suture; scl = scutellum; sclf = scutellar foramen; scp = scape; sct = scutum; scts = scutal setae; sp 2 = mesothoracic spiracle; sp3 = metathoracic spiracle; stn 1 = prosternum; stn1s = prosternal setae; stn2 = basisternum or mesosternum; stn3 =metasternum; $tar_3 = tarsus$ of metathoracic leg; teg = tegula; tegs = tegular setae; tdgt = tarsaldigitules; ti3 = tibia of metathoracic leg; tibs= tibial spur; tp = triangular plate; tr 3 = trochanter of metathoracic leg; vmcr = ventral midcranial ridge; vse = ventral simple eye.

Abdomen

Segments I–VII: tergites (*at*) I–IV unsclerotised, V–VII slightly sclerotised; sternites (*as*) I–IV weakly sclerotised, V and VII fairly well sclerotised. Caudal extension of segment VII (*ce VII*) tapering, weakly sclerotised, with 11–17 (12) pleural *fs* and 2 or 3 *hs*. Dorsal abdominal setae (*ads*) (total across segment): I 16–24 (20) *fs*; II 16–24 (20) *fs*; II 16–24 (20) *fs*; III 4–16 (10) *fs* + 2 *hs*; IV 6–20 (13) *fs* + 2 *hs*; V 2–14 (8) *fs* + 2 *hs*; VI 2–18 (11) fs + 2–4 *hs*; VII 6–10 (8) *fs* + 2–6 (4) *hs*. Pleural setae: dorsopleural setae (*dpls*) (on each side): I 2 or 3 *fs* and generally 1 *hs*; II 1–4 (2) *fs* + 1 *hs*; III 1–4 (2) *fs* + 1 or 2 *hs*; IV 2–4 *fs* + 1 *hs*; V 2–6 (3) fs + 1–2 *hs*; VI 2–4 (3) *fs* + 1–4 (2) *hs*, and VII 1–3 *fs*

+ 1 or 2 hs. Ventropleural setae (vpls) (on each side): I 2–8 (4) fs + 0 or 1 hs; II 1–4 (2) fs; III 2-6 (3) fs + 0 or 1 hs; IV 1-6 (3) fs + 1 hs; V 1-4 (2) + 1 or 2 hs; VI 1-3 (2) fs + 1 or 2 hs; VII 1–4 (2) fs + 1 or 2 hs on VII. Ventral abdominal setae (totals): I 10–21 (16) fs; II 10–22 (17) fs; III 12–16 (14) fs + 2 hs; IV 10–16 (12) fs + 2–4 hs; V 7–16 (11) fs + 2-6 (3) hs; VI 4-10 (8) fs + 2-4 hs; and VII 4-10 (8) fs + 2-6 (4) hs. Segment VIII: tergite (at) and sternite (as) sclerotised; tergite sometimes with a pair of short fs dorsal abdominal setae (ads); sternite without ventral abdominal setae; caudal extension of segment VIII (ce VIII) weakly sclerotised, semi-circular lobe; cicatrix absent; with 0-2 hs ventral abdominal setae; glandular pouches absent, but with about 4 long hs in this position. Genital segment: anus present just anterior to penial sheath on dorsal surface, 27-42 (37) µm wide. Penial sheath (ps) long, anteriorly with parallel sides, posteriorly with a pointed apex; covered by a membranous extension from segment IX; penial sheath with a group of small sensilla near apex; also with 10-17 (14) fs and 8–21 (14) minute hs penial sheath setae (gfs); fs mainly on basal portion of penial sheath margin. Penial sheath 244-310 (267) µm long and 63-111 (75) µm wide at base; ratio of total body length to penial sheath length about 1:4. Basal rod (bra) distinct, positioned just posterior to basal membranous area of aedeagus, 248-315 (291) µm long and 15–19 µm wide; aedeagus (aed) with parallel sides; 178–296 (233) µm long, lying within penial sheath, membranous extension of aedeagus absent.

Comments: the membranous area of the scutum of *C. japonicus* has 10-14 (12) *fs* and 6-10 (8) *hs* scutal setae (*scts*) which is fewer than the 45 reported for *C. ceriferus* by Gimpel *et al.* (1974). On the other hand, *C. cirripediformis* apparently has about 23 scutal setae and glandular pouches and associated setae, that are absent on both *C. japonicus* and *C. ceriferus* (Gimpel *et al.*, 1974). The penial sheath has unusual fleshy setae (*gfs*) on the margins of the basal part, a character shared with *C. cirripediformis*. Fleshy setae are absent from the penial sheaths of *W. berliniae* and *Waxiella* sp. (Giliomee, 1967). Our description is similar to the draw- ings of *C. japonicus* penial sheath by Borchsenius (1957, p. 30, figs 58, 59).

SECOND-INSTAR MALE NYMPH (Fig. 2)

Described from 6 specimens in good condition.

Unmounted material: body oval, lightly convex dorsally, brown.

Mounted material: body elongate oval; 924–1147 (1058 μ m) long; 551–671 (626 μ m) wide; anal cleft short.

Dorsum: derm membranous. Preopercular pores absent, other dorsal pores not seen. Tubular ducts each duct with cup-shaped invagination, $3-5 \ \mu m$ wide; outer ductule 16–25 (21 μ m) long; inner ductule 14–19 (17 μ m) long, terminal gland, 3–6 (5 μ m) wide; ducts distributed in a single row along body margin, but absent from posterior 1/5 of margin; 6–11 (9) anteriorly between eyespots, 2–6 (4) between eyespot and anterior spiracular furrow, 4–8 (6) anteriorly between anterior and posterior spiracular furrows, and 5–7 posterior each posterior spiracular area. Anal plates each subtriangular, broad, with inner margins slightly diverging; dimensions of each plate 26–39 (31) μ m broad; anterior margin 35–46 (41) μ m long; posterior margin 42–55 (47) μ m inner margin 49–64 (58) μ m long; each plate with 1 posterior margin seta, 25–32 (28) μ m long, 2 inner margin setae, anterior 9–13 (10) μ m long. All anal plate setae slightly spinose. Anogenital fold with 3 pairs of anterior margin setae and 1 pair of lateral margin setae. Anal ring with 6 setae.

Margin: marginal setae in a single line, each curved and setose, and 9–10 μ m long; with 5–6 anteriorly between eyespots, and, on each side, 2 between eyespot and anterior spiracular area, 2 between anterior and posterior spiracular area, and 6 between posterior spiracular area and anal cleft; with two longer setae on at apex of each anal lobe, 22–30 (26) μ m and 16–22 (19) μ m long respectively. Stigmatic clefts shallow, each with 3 short, conical stigmatic spines, median spine longest and slightly set onto dorsum, 13–19 (18 μ m) long and 4–7 (6 μ m) wide at base; lateral setae 9–12 (10 μ m) long and 4–6 μ m wide at base.





Where A = antenna and interantennal setae; B = spiracular quinquelocular disc-pore; C = ventral microduct with cruciform pore; D = metathoracic leg; E = abdominal ventral seta; F = ventral dermal spinules; G = dorsal view of anal plates (left) and view of ano-genital fold (right); H = view of stigmatic cleft; I = dorsal tubular duct; J = marginal seta.

Venter: derm membranous, segmentation obscure; minute dermal spinules most frequent around anal cleft. Ventral microducts, each less than 1µmwide, sparsely distributed in a submarginal band. Ventral setae bristle-like, each 1–2 µm long, present in submarginal and submedial rows on abdominal segments. Two pairs of interantennal setae present, shorter pair 9–19 (15 µm) long and longer pair 26–39 (33 μm) long respectively. Antennae 6-segmented, each 107-126 (117) μm long, third segment longest. Spiracles: all peritremes 10-13 (11)µmwide; spiracular disc-pores quinquelocular, each about 3 μ m wide, with 8–12 (10) forming a narrow band about 1 pore wide from each spiracle to body margin. Legs well developed, without tibiotarsal scleroses; claws without denticles; claw digitules unequal, one broad apically and 12-17 (15) µm long, other slender, with a knobbed apex and 12-17 (15) µm long. Coxae: 43-48 (46) µm long, 28-39 (34) µm wide; with 4 ss, longest 25-38 (29) µm long. Trochanter + femur: 71–78 (75) µm long, 23–29 (25) µm wide; trochanter with 1-3 (2) ss, longest 29-42 (36) µm long; femur with 1 or 2 short ss. Tibia 46-51 (49) μm long, 12–14 (13 μm) wide; with 1 or 2 ss. Tarsus 39–42 (41) μm long, 10–13 (12) μ m wide; with 2 or 3 ss, longest 9–16 (12) μ m long; tarsal digitules each 25–41 (29) μm long.

Comments: the only described 2nd-instar male nymph of a *Ceroplastes* species is that of *C. sinensis* (Qin & Gullan, 1994). The 2nd-instar male nymphs of *C. japonicus* and *C. sinensis* are very similar, the main difference appearing to be the number of spiracular disc-pores, with an average of 10/band in *C. japonicus* and 6/ band in *C. sinensis*.





Where: A = dorsal dermal spinules; B = ventral dermal spinules, and C = dorsal (left) and ventral (right) views of posterior end of abdomen. And where: ab II–VIII = abdominal segments II–VIII; ads = dorsal abdominal setae; asp = anterior spiracle; avs = ventral abdominal setae; ce VII= caudal extension of segment VII; ce VIII = caudal extension of segment VIII; h = head; ps = penial sheath; psp = posterior spiracle; sdp = spiracular discpore; vpls = ventropleural setae; vts = ventral thoracic setae; wb = wing bud

PREPUPA (Fig. 3)

Described from 9 specimens in good condition.

Mounted material: body elongate, narrowest at head, widest across abdomen, length 775–1132 (855 m), width across abdomen 402–566 (469 μ m). Division into head, thorax and abdomen not clear. Segmentation fairly distinct on abdomen.

Head: lacking mouthparts and simple eyes. Antennae fairly well sclerotised, elongate, each 174–211 (188 μ m) long, with 9 poorly defined segments; antennal length to body length ratio 1:4.45–5.36 (4.55); antennal setae: IX: with 4 minute fleshy setae (*fs*); VIII and VII each with 1 minute *fs*. With 2 pairs of minute interantennal setae ventrally plus 2 minute setose dorsal head setae, each 3–4 μ m long.

Thorax: prothoracic legs directed anteriorly, only reaching to base of each antenna; other pairs directed posteriorly; claws and digitules extremely reduced; metathoracic legs each 120–196 (160 μ m) long. Wing buds each 155–207 (183) μ m long, 70–104 (92) μ m wide, ratio of length to width 1: 0.45–0.50. Spiracles: width of peritremes 15–20 (17) μ m, each with 6–12 (8) spiracular disc-pores near atrium; pores mainly quinquelocular and about 4 μ m wide, but some with many more loculi. Setae: with 2 minute thoracic setae postero-laterally to each procoxa + 1 minute fleshy ventropleural seta on mesothorax.

Abdomen: segmentation fairly well defined; minute dermal spinules most dense on posterior segments; dermal spinules with a different pattern on dorsum with respect to

venter. Setae: pairs of minute dorsal abdominal setae (*ads*) present medially on segments IV–VII, each about 4 μ m long; with a pair of ventral abdominal setae (*avs*), each 6 μ m long, on segments II–VII. Margins with minute ventropleural setae (*vpls*), each 4–12 (7 μ m) long, segmentally arranged, with a single seta on each side of segments I–IV and 2 or 3 fleshy setae on segments V–VIII. Caudal extension VII (*ce VII*) sclerotised, 58–112 (94) μ m long, 41–83 (59) μ m wide, with 2 apical hair-like setae, 16–23 (20) μ m long and 9–19 (15) μ m long respectively, 1 short seta 4–7 (6) μ m long + 1 dorsopleural seta at base of ce VII, 6–9 (7) μ m long. Caudal extension VIII (*ceVIII*) poorly developed and slightly sclerotised, with 3 minute setae. Penial sheath sclerotised, roundly triangular in shape, 55–78 (69) μ m long and 55–78 (69) μ m wide, ratio of length to width 1:1, with 2 ante-anal setae on segment VIII, and 2 pairs of minute setae dorsally on penial sheath.

PUPA (Fig. 4)

Described from 8 specimens in good condition.

Mounted material: elongate oval, tapered anteriorly, widest across abdomen, length 819-983 (926) μ m, maximum width across abdomen 343-417 (376) μ m. Derm membranous. Division into head, thorax and abdomen not clear. Segmentation not well defined, most distinct on abdomen.

Head: fairly well sclerotised, sometimes dorsal derm with circular sclerotisation, lacking mouthparts and simple eyes. Antennae slightly sclerotised, elongate; directed postero-laterally, each antenna with 9 fairly well-defined segments, 333–429 (363 μ m) long; antennal length to body length ratio 1:2.29–2.46 (2.55); antennal setae distribution: IX: 6; VIII: 1, VII: 1; each about 3 μ m long; segments IX and VIII each with 1 *fs* about 3 μ m long. With 1 or 2 minute dorsal setae anteriorly on head, each about 3 μ m long; ventrally with 2 very short head setae.

FIGURE 4. Ceroplastes japonicus Green, pupa.



Labels as fig 3, except A = pattern of dorsal head sclerotisation; dpls = dorsal pleural setae.

Thorax: legs well developed, each with clear segmentation; prothoracic legs directed anteriorly, extending anteriorly to head; other pairs directed posteriorly; metathoracic legs each 389–499 (435) μ m long. Wing buds each 377–429 (404) μ m long, 118–140 (128) μ m wide; ratio of length to width 1:0.31–0.33 (0.32). Spiracles: width of peritremes 14–20 (18) m, each anterior spiracle with 10–16 (13) spiracular disc pores near atrium, each about 4 μ m wide, and each posterior spiracle with 7–10 (9) disc pores; number of loculi highly variable. Setae: ventrally with 1 minute seta near each coxa.

Abdomen: segmentation fairly well defined. Setae: with 2 pairs of minute dorsal abdominal setae (*ads*), each 4 µm long, on segments IV–VII; 1 minute dorsopleural seta (*dpls*) on segments V–VII; 1 or 2 minute ventral abdominal setae (*avs*) on segments III–VII, each about 1.5 µm long, + 1 or 2 ventropleural setae (*vpls*) on each side of segments II–VII, each seta 4–8 (7) µm long; ante-anal setae not detected. Caudal extensions on segment VII (*ce VII*) well developed, each 52–87 (67) µm long and 29–39 (34) µm wide at base; each with 2 spinose setae, 19–22 (21) µm and 10–19 (14) µm long respectively, + 1 minute hair-like seta, 4–7 (6) µm long; each *ce VII* also with 1 *hs* ventrally near base, 4–6 µm long. Caudal extensions on segment VIII (*ceVIII*) poorly developed, with 3 *hs* and 1 *fs* minute setae. Penial sheath (*ps*) elongate-triangular, sclerotised, 110–123 (116) µm long, 61–68 (142) µm wide at base, ratio of length to width 1 : 0.55; dorsally with 3 pairs of *hs*. Anal opening at base of penial sheath, 19–22 (20 µm) wide.

Conclusions :

With the present descriptions and illustrations, all the instars of *C. japonicus* are now described and illustrated. Good descriptions and illustrations of the adult female are provided by De Lotto (1969), Tang (1991) and Pellizzari & Camporese (1994); the 1st-instar nymph, 2nd- and 3rd-instar female nymphs have been described by Camporese & Pellizzari (1994) and further descriptions and illustrations of some female instars and the 1st- instar nymph can be found in Xie *et al.* (2006). Affinities between species will be discussed together with *C. rusci* (Section II).

SECTION II

DESCRIPTION OF THIRD-INSTAR FEMALE NYMPH AND MALE INSTARS OF *CEROPLASTES RUSCI* L. (HEMIPTERA: COCCOIDEA: COCCIDAE), WITH A DISCUSSION OF ITS AFFINITIES WITH OTHER *CEROPLASTES* SPECIES BASED ON THE ADULT MALE

Paper to be submitted as :

Rainato A., Pellizzari G.. Descriptions of male nymphal instars and adult male of *Ceroplastes rusci* L. (Hemiptera: Coccoidea: Coccidae), with a discussion of its affinities with other *Ceroplastes* species based on the adult male.

Introduction

In the last years several studies have been devoted to the genus *Ceroplastes* with the aim of clarifying the native areas of many species, their present distribution, their morphology, (mainly of young stages and males) and with revision and description of new species (Pellizzari & Camporese, 1994; Camporese & Pellizzari, 1994; Qin & Gullan, 1994; Qin & Gullan, 1995; Qin *et al.*, 1994; Qin *et al.*, 1998; Wakgari & Giliomee, 1998; Ben-Dov *et al.*, 2000; Rainato & Pellizzari, 2008; Peronti *et al.*, 2008). The genus has presently a worldwide distribution, due also to human activity, and comprehends several notorious plant pests. Among them, the fig wax scale *Ceroplastes rusci*, credited as native from Afrotropical region (Qin *et al.*, 1994; 1998), is widespread in the Palearctic Region and also recorded in some African countries, in the Neotropical Region (Argentina, Antigua, Brazil) (Granara de Willink, 1999) and in very few Oriental countries (Irian Jaya and Vietnam) (Williams & Watson, 1990; Vu *et al.*, 2006).

C. rusci is widely distributed throughout costal areas of Mediterranean countries, and is the ancient *Ceroplastes* species recognized in this area since Theophrastus times $(370 \text{ B.C.} - 285 \text{ B.C.})^1$ (Silvestri & Martelli, 1908). *C. rusci* has a wide range of host plants, and is occasionally a pest in Citrus groves and tropical fruit orchards, but in fact it is almost always recorded on the same common plants of the Mediterranean maquis: *Nerium oleander, Pistacia lentiscus P. terebinthus, Myrtus communis, Ficus carica.* Apparently fig is a favourite host and on this plant heavy infestations are quite common (Bodkin, 1927; Khasawinah & Talhouk, 1964). This paper was stimulated by the heavy infestations of *C. rusci* on fig cultivations presently occurring in Messinia, District of Kalamata (Greece) where fig is an important fruit crop, the annual fig production reaches roughly 4000 tons/year and dried figs are usually exported towards American and European markets.

In Messinia infestation by *C. rusci* are kept under an unsatisfactory control by chemicals. Successful chemical control needs a proper timing and the correct identification of the stages present on the host plant is essential.

¹ According to Silvestri & Martelli (1908) Theophrastus described the symptoms of C. rusci on fig in his book "De causis plantarum".

In the *Ceroplastes* species the females goes through four development stages. Teneral adult females are macroscopically indistinguishable from third instars and also the identification of the different instars is unreliable, in absence of a detailed microscopical description and illustrations of mounted specimens to which refer. Failures in chemical control could be ascribed to lack of knowledge about scale instars present on the treated trees. These reasons stimulated the description of young instars of *C. japonicus* (Camporese & Pellizzari, 1994) and *C. destructor* (Wakgari & Giliomee, 1998)

Despite its large distribution and pest status in many countries, nymphal female instars of *C. rusci* have not yet been described in a proper way: previous old descriptions by Silvestri & Martelli (1908) or or Khasawinah & Talhouk (1964), are useless. Moreover, adult male and male instars are undescribed.

CEROPLASTES RUSCI L.

Coccus rusci Linnaeus, 1758: 456. *Ceroplastes rusci* (Linnaeus); Signoret, 1872b: 35.

Material examined. **GREECE**: Kalamata, on *Ficus carica*, 28.vi.2007, G. Pellizzari: 21 third instar females, 15 adult males, 10 second-instar males, 10 prepupae, 8 pupae.

ADULT MALE (Fig 1)

Described from 8 males in fairly good condition; details checked on remaining specimens. Male tests not available for detailed descriptions.

Mounted material: total body length 1296-1386 (1386) μ m; maximum width across triangular plates 298-432 (390) μ m. Body and appendages covered with numerous setae, mostly fleshy setae (*fs*); hair-like setae (*hs*) fewer. Dermal pores entirely absent. Abdominal glandular pouches and associated setae present. Caudal extensions present on abdominal segments VII and VIII.

Head

Bluntly triangular in dorsal viewlength from apex to pronotal ridge (prnr) 163-185 (175) µm, width across genae 196-237 (215) µm. Median crest (mc) showing polygonal reticulations; dorsal head setae (*dhs*): with 6-10 (8) *fs* and 6-14 (9) *hs*. ventral midcranial ridge (*vmcr*) strong and well developed, extending with short lateral arms (*lmcr*) from behind the level of posterior margin of ocular sclerite (*ocs*); midcranial ridge dorsally absent. Ventral midcranial ridge setae (*vmcrs*) not seen on the available specimens. Genae (g) sclerotised, with polygonal reticulations and 22-30 (26) fs and 8-12 (9) hs genal setae (gs) on each side. Simple eves (se) two pairs subequal in size, rounded, in some specimen corneas of ventral simple eye (vse) stretched sagittally; dorsal eyes (dse) situated on head apex, each 36-42 (39) µm wide; ventral eyes (vse) on posterior ventral area of head, each 41 µm wide. Ocelli (o) situated laterally, each 15-20 (17) um wide, lying just anterior to postocular ridge (pocr); interocular ridge (ior) absent. Ocular sclerite (ocs) fairly well sclerotised, with polygonal reticulations throughout. Preocular ridge (procr) distinct but short; postocular ridge (pocr) strongly developed. Dorsal ocular setae (dos) 4-5 fs and 1 hs. Ventral head setae (vhs) 15-24 (21) fs, mainly situated antero-laterally to ventral simple eyes. Preoral ridge (pror) fairly-well developed and U shaped, about 21 µm of total length, detected in only 2 specimens. Cranial apophysis (ca) fairly well defined, with bifurcate apex, 15-33 (23) µm long.

Antennae: ten-segmented and filiform, with short, stout fleshy setae (*fs*); antennae 536-641 (560) µm long; shorter than half body length (ratio of total body length to antennal length 1: 0.41-0.46. average 0.44), shorter than posterior leg (ratio of posterior leg length to antennal length 1:0.17-0.21, average 0.20) and longer than penial sheath (ratio of penial sheath to antennal length1:1.56-1.68, average 1.63). Scape (*scp*) subrectangular in shape, each 32-38 (36) µm long, 33-44 (39) µm wide; with 3 *hs*. Pedicel (*pdc*), each 33-42 (38) µm long, 28-36 (32) µm wide; with 5-6 *fs* and 4-5 *hs*, and with polygonal reticulations on distal 1/3. Segment III long and club-shaped, but variable in shape, 5-3 (3,35) times longer than wide; each 70-75 (72) µm long, 15-25 (21) µm maximum width, with 9-13 (11) *fs* and 2 *hs*. Segments IV-IX cylindrical, each

Figure 1. Ceroplastes rusci L., adult male.



Where, A = antennal segments: A_I = segments I-III; A_{II} = segment X; B = body setae: fleshy seta (B_I), hair seta (B_{II}); C = metatarsus and claw; D = dorsal and ventral view of the genital segment; $E = dorsal (E_I)$ and ventral (E_{II}) view of polygonal reticulations on head capsule. And where abr = antennal bristles; ads = dorsal abdominal setae; aed = aedeagus; $ams_{3}s =$ ante metaspiracular setae; amss = anterior metasternal setae; a_{NIII} = abdominal segment II-VIII; $at_I - at_{VIII}$ = abdominal tergite I-VIII; avs = abdominal ventral setae; bra = basal rod of aedeagus; c = claw; cb = coxal bristles; ccx = costal complex of wing veins; cdgt = clawdigitules; ce_{VII} = caudal extension of segment VII; ce_{VIII} = caudal extension of segment VIII; $cx_3 = coxa$ of methathoracic leg; dos = dorsal ocular setae; dpls = dorsopleural setae; dse = dorsal simple eye; dss = dorsal spiracular setae; eps_2 = mesepisternum; f = furca; fm₃ = femur of metathoracic leg; g = genae; gfs = genital fleshy setae; gs = genal setae; mc = median crest; mr = marginal ridge; o = ocellus; pa = postalare; pdc = pedicel; pepcv = proepisternum+cervical sclerite; $pms_2 = post$ mesospiracular setae; pmss = posterior metasternal setae; $pn_2 =$ mesopostnotum; pna = postnatal apophysis; pocr = preocoular ridge; pra = prealare; prn = lateral pronotal sclerite; prnr = pronotal ridge; prsc = prescutum; ps = penial sheath; pscr =prescutal ridge; pscs = prescutal suture; scl = scutellum; sclf = scutellar foramen; scp = scape; sct = scutum; scts = scutal setae; sp_2 = mesothoracic spiracle; sp_3 = metathoracic spiracle; stn_1 = prosternum; stn_1s = prosternal setae; stn_2 = basisternum or mesosternum; tar_3 = tarsus of metatxhoracic leg; teg = tegula; tegs = tegular setae; tdgt = tarsal digitules; ti₃ = tibia of metathoracic leg; tp = triangular plate; tr_3 = trochanter of metathoracic leg; vmcr = ventral midcranial ridge; vse = ventral simple eye.

[...] about 16-26 (20) wide; lengths (μ m): IV: 99-110 (105); V: 83-93 (87); VI: 64-73 (67); VII: 65-73 (67); VIII: 46-53 (41); IX: 41-62 (48); setae distribution: IV 18-27 (22), V 18-20 (19), VI 15-26 (19), VII 12-28 (19), VIII 12-15 (14), IX 13-15 (14) *fs* respectively; *hs* absent on the examined specimens. One antennal bristle (*abr*) distinctly larger than *fs* present on segments VIII & IX, 30-38 (33) μ m and 32-41 (35) long respectively. Segment X 55-70 (61) μ m long, 16-29 (21) μ m wide; with 8-9 *fs*, 1-2 hair setae (*hs*), 3-5 (4) antennal bristles (*abr*), each 41-58 (48) μ m long, and 3 subapical capitatae setae (caps), each 41-58 (48) μ m long; with apex constricted.

Thorax

Prothorax: membranous; pronotal ridges (*prnr*) well developed, interrupted by weak sclerotisation dorsomedially; pronotal sclerite (*prn*) present, without lateral pronotal setae (*lpns*). Median pronotal setae (mpns) absent; post-tergite (*pt*) apparently absent. Prosternum (*stn*₁) lightly sclerotised; bounded posteriorly by a strong transverse ridge, with median ridge well developed; with total of 6-12 (10) *fs* prosternal setae (*snt*₁*s*). Anteprosternal setae (*astn*₁*s*) 4-6 fs. Antemesospiracular setae (*ams*₁*s*): 3 pairs.

Mesothorax: mesoprephragma with pronounced ventral emargination. Prescutum (prsc) 85-122 (105) µm long; 170-196 (185) µm wide; heavily sclerotised, showing regular polygonal reticulations; anterior margin curved; laterally bounded by heavily sclerotised prescutal ridges (prcr) and posterior margin of prescutum bounded by the prescutal suture (pscs). Scutum (sct): with median membranous area trapezoidal or sub-rectangular, slightly wider posteriorly; 67-93 (78) µm long; 174-207 (187) µm wide; scutal setae (scts): 10-12 (11) fs and 6-10 (8) hs. Rest of the scutum heavily sclerotised and showing polygonal nodulations, but without setae. Scutellum (scl) 44-55 (48) μm long, 185-207 (195) μm wide; tubular, with oval ventral foramen (sclf) small; without either a scutellar ridge (sclr) or scutellar setae (scls). Prealare (pra) and triangular plate (tp) well developed. Mesopostnotum (pn_2) well developed; postnotal apophysis (*pna*) and postalare well developed and strongly sclerotised. Postalare (*pa*) without nodulation and without postalare setae (pas). Mesopostphragma with deep emargination. Mesopleural apophysis (pla_2) and mesopleural wing process (pwp_2) well developed. Basalare (bas) well developed. Subalare (sa) present. Mesepisternum (eps₂) well sclerotised, with fairly defined nodulations; subepisternal ridge (ser) well developed. Mesepimeron (epm_2) undetected and probably undeveloped. Lateropleurite (lpl) narrow. Basisternum (stn₂) 122-148 (136) µm long, 246-270 (266) µm wide; with strong medial ridge and bounded anteriorly by strong marginal ridge (ma) and posteriorly by a strong precoxal ridge (pcr_2) ; without setae; furca (f) well developed, narrow waisted, with arms divergent and extending about ³/₄ way to marginal ridge anteriorly. Mesothoracic spiracle (sp_2) with well developed peritreme; width of peritreme 16-22 (20) µm; post-mesospiracular setae (pms): 31-36 (34) fs, arranged in a band across segment between spiracles. Tegula (*teg*) present, with 4-6 (5) *hs* tegular setae (*tegs*).

Metathorax: suspensorial sclerites (*ss*) absent. Metapostnotum (*pn*₃) undetected and probably absent; metatergal setae (*mts*) were not observed on the available specimens; dorsospiracular setae (*x*) 4-6 *fs*. Metapleural ridge (*plr*₃) reduced and short, with metapleural ridge wing process (*pwp*₃) absent. Metasternum weakly sclerotised; anterior metasternal setae (*amss*) 15-20 (18) *fs*; posterior metasternal setae (*pmss*) 18-25 (20) *fs*. Metepisternum (*eps*₃) membranous, with 5-8 (6) *fs* postmetaspiracular setae (*eps*₃*s*); precoxal ridge (*pcr*₃) well developed, with metepimeron (*epm*₃) undetected and probably reduced or absent. Metathoracic spiracle (*sp*₃) similar to mesothoracic one; width of peritreme 17-25 (21) µm. *Wings*: hyaline; rather short and comparatively broad; 819-938 (884) µm long and 343-447 (405) µm wide; ratio of width to length 1:2.1-2.39 (2.18); ratio of total body length to wing length 1:0.63-0.68 (0.67); alar lobe (*al*) and alar setae (*als*) absent. Hamulohalterae (*h*) absent.

Legs: long and slender, metathoracic leg longest, prothoracic leg subequal to the mesothoracic; total lengths (μ m): I: 592-629 (615); II: 592-659 (622); III: 636-928 (733) μ m; ratio of hind leg to total body length 1:1.5-2.0 (1:1.8). Coxae (*cx*): I: 52-67 (61); II: 63-81 (69); III: 67-85 (76) μ m long; setae of coxa III: 20-22 (21) *fs* and 4-6 (5) *hs*. Trochanter (*tr*) + femur (*fm*) lengths (μ m): I: 185-233 (210); II: 185-204 (194); III: 166-211 (193); maximum widths: I: 30-46 (38); II: 31-49 (39); III: 34-51 (40); ratio of maximum width to length of hind femur 1:4.1-4.9 (4.8); with 2 oval sensilla on each side of trochanter (*tcp*). Trochanter III with 10-18 (13) *fs* and 2-4 *hs*; femur III with 23-31 (28) *fs* and 4-8 (6) *hs*. Tibia (*ti*): I: 252-274 (265) II: 229-281 (260); III: 263-296 (283) μ m long (ratio of length to width of tibia III 1:14-16 (15)); tibia III with 40-51 (47) *fs*; 8-12 (10) *hs*, more numerous near apex; with one tibial spur (*tibs*) on the inner margin, 22-26 (24) μ m long. Tarsi one-segmented: I: 93-96 (94); II: 97-102 (100); III: 99-103 (101) μ m long (ratio of length of tibia III to length of tarsus III 1:0.37-0.35 (0.36); tarsus III with 20-22 (21) *fs*, 8-10 (9) *hs*; tarsal campaniform pores (*tcp*) absent; tarsal digitules (*tdt*) capitate, each 22-29 (26) μ m long. Claws (*c*) short,

slightly curved and pointed, with minute claw denticle (*cd*): length III 19-23 (21) μ m, claw digitules (cdt) capitate, each 22-26 (24) μ m long.

Abdomen

Segments I-VII: tergites (*at*) I-II mainly unsclerotised, III-VII slightly sclerotised; sternites (*as*) II-VII fairly well sclerotised, with sclerotisation concentrated on the median part of each segment. Caudal extension of segment VII (ce_{VII}) very prominent and tapering, fairly well sclerotised ventrad, usually *hs* pleural setae situated near apex longer and spine-like. Dorsal abdominal setae (*ads*) (totals): I-II absent; III 4 *fs*; IV 4 *fs* + 4 *hs*; V 4-8 *fs* + 4 *hs*; VI 4 fs + 4-8 *hs*; VII 0-2 *fs* + 4 *hs*. Pleural setae: dorsopleural setae (*dpls*) (on each side): I absent; II 2 *fs*; III 2 *fs*; IV 2-4 *fs* + 2 *hs*; V 1-4 (2) fs + 2 *hs*; VI 2-4 *fs* + 2 *hs*, and VII 4-6 *fs* + 2-3 *hs*. Ventropleural setae (*vps*) (on each side): I absent; II 4 *fs* + 1 *hs* ; III 2 *fs*; IV 2 fs + 2 *hs*; V 2-4 *fs* + 2-4 *hs* ; VI 2-4 *fs*; VII 7-10 (9) *fs* + 2 *hs* on VII. Ventral abdominal setae (*avs*) (totals): II 20-2 (23); III 16-20 (18) *fs* + 2 *hs*; IV 8-12 *fs* + 2 *hs*; V 6-10 fs + 2 hs; VI 6-10 (8) *fs* + 2 *hs*; and VII 4-6 *fs* + 2 sometimes 4-6 *hs*.

Segment VIII: tergite (*at*) and sternite (*as*) sclerotised; dorsal abdominal setae (*ads*) (total): 4 *hs*; dorsopleural setae (*dpls*) (on each side): 2 fs + 2 hs; ventropleural setae (*vps*) (on each side): 2-4 *fs* + 2 *hs*; sternite without ventral abdominal setae (*avs*); caudal extension of segment VIII (*ce_{VIII}*) weakly sclerotised, forming a prominent semi-circular lobe; dorsal cicatrix (*c*) present, with nodulations undetected; glandular pouches (*gp*) present, which contain 2 long flagellate setae, 138-177 (163) µm long, whose protruding part is about 5 times as long as the section within the pouch. Small tergal plate of segment IX present; with 2 *fs* ante anal setae. Posterior margin with 4-5 fs and 3 hs on each side.

Genital segment: penial sheath (*ps*) long, anterior part with parallel sides, posteriorly with a pointed apex, this produced into a small and finger-like membranous extension, 57-87 (69) μ m long; ventrally covered by a membranous extension from segment IX; anal opening (*an*) less than 1,5 μ m wide; penial sheath with a group of small sensilla near apex; also with 16-28 (23) *fs* and 12-20 (15) minute *hs* penial sheath setae (*gts*)

mainly ventrally on margins; *fs* absent on the apical portion of penial sheath. Penial sheath 344-381 (361) μ m long and 80-84 (82) μ m wide at base; ratio of total body-length to penial sheath length about 1:3.7. Basal rod (bra) distinct, positioned just posterior the basal membranous area (*bma*) of the aedeagus, 145-188 (166) μ m long and about 19 μ m wide; aedeagus (*aed*) with parallel sides; 203-222 (213) μ m long, lying within the penial sheath.

Comments

The following comparison of some morphological characters of *Ceroplastes* and *Waxiella* males includes data of *C. cirripediformis* and *C. ceriferus* from Gimpel *et al.* (1974), of *C. japonicus* from Rainato & Pellizzari (2008) and data of *W. berliniae* and *Waxiella* sp. from Giliomee (1967).

Head: ventral midcranial ridge setae are absent in *C. rusci* male; there are 4 hair setae in *C. japonicus* male, and they are present in *Waxiella berliniae* and *Waxiella* sp. males. In *C. rusci* male the antennal segment III is as long as in *Waxiella* spp. males, whereas in *C. japonicus* and *W. berliniae* males it is significantly shorter.

Thorax: in *C. rusci* male the scutum has 10-12 (11) fleshy setae and 6-10 (8) hair-like setae scutal setae. This number is about the same of *C. japonicus* male and close to that of *C cirripediformis* male. On the other hand, *C. ceriferus* presents 45 scutal setae. Metatergal setae were not found, but this character is difficult to detect due to the sclerotisation of this area. This feature is shared only with *C. cirripediformis*.

Abdomen: glandular pouches are present in *C. rusci* male: this character is shared with *Waxiella berliniae*, *Waxiella* sp. and *C. cirripediformis* males, but not with *C. ceriferus*. Caudal extensions of segment VIII do not present any minute seta in *C. rusci* male; they are instead present on *C. japonicus* and *C. ceriferus*. Several fleshy setae occur on the basal part of the penial sheath margin in *C. rusci* male, a character shared with *C. cirripediformis*, *C. japonicus* and *C. ceriferus* males, even if with a different distribution. The number of fleshy setae of *C. rusci* male is slightly higher

than that reported for *C. japonicus* male. Fleshy setae are absent from the penial sheaths of *W. berliniae* and *Waxiella* spp..

SECOND INSTAR MALE NYMPH (Fig. 2)

Described from 10 specimens in good condition. Details checked on remaining specimens.

Unmounted material: body oval, lightly convex dorsally, brown in colour.

Mounted material: body elongate oval; 1043-1267 (1150 μ m) long; 581-745 (654 μ m) wide; anal cleft short.

Dorsum: derm membranous. Preopercular pores and other dorsal pores absent. Tubular ducts in a single row along body margin, except posterior 1/5 of margin; each duct with external cup-shaped invagination: 4-6 (5 μ m) wide; outer ductule: 19-24 (23 μ m) long; inner ductule: 20-23 (21 μ m) long, terminal gland: 3-6 (5 μ m) wide; distribution of ducts: 11-13 between eyespots, 5-6 between eyespot and anterior spiracular furrow, 6 between anterior and posterior spiracular furrows, 6 posterior each spiracular area. Anal plates each subtriangular, broad, with inner margins slightly diverging; dimensions of each plate 29-45 (37) μ m broad; anterior margin 39-49 (39) μ m long; posterior margin 45-54 (49) μ m, inner margin 55-58 (56) μ m long; each plate with 1 posterior margin seta, 26-30 (26) μ m long, 2 inner margin setae, 16-20 (18) μ m and 19-25 (21) μ m long respectively, with one apical seta 16-19 (18) μ m long. All anal plate setae slightly spinose. Anogenital fold with 3 pairs of anterior margin setae and 1 pair of lateral margin setae. Anal ring with 6 setae.

Margin: marginal setae arranged in a single marginal line, each setose and 7-10 (8) μ m long, sometimes curved; with 8-9 between eyespots, and, on each side, 2 between eyespot and anterior spiracular area, 3-4 between anterior and posterior spiracular area, and 8-10 (9) between posterior spiracular area and anal cleft; with one longer setose seta on each anal lobe, each 29-36 (32) μ m long. Stigmatic clefts shallow, each with 3

short, conical stigmatic spines, median spine longest and slightly set onto dorsum, 13 μ m long and 6 μ m wide at base; lateral setae 9-12 (10) μ m long and 4 μ m wide at base.

Venter: derm membranous, segmentation obscure; minute dermal spines most frequent around anal cleft. Ventral microducts, each about 1,5 µm wide, sparsely distributed in a submarginal band. Ventral setae bristle-like, each 1-2 µm long, present in submarginal and submedial rows on abdominal segments. Two pairs of interantennal setae: shorter pair 4-9 (7 µm) long and longer pair 23-37 (34 µm) long respectively. Antennae 6-segmented, each 131-151 (141) µm long, third segment longest; dimensions : segment I: 22-25 (23) µm long and 25-30 (27) µm wide; II: 16-19 (17) μm long and 19-25 (21) μm wide; III: 39-46 (43) μm long and 16-17 (16) μm wide; IV: 12-15 (13) µm long and 12-15 (13) µm wide; V: 13-16 (15) µm long and 10-12 (11) µm wide; VI: 23-30 (27) µm long and 9-13 (11) µm wide; antennal setae: segment I: 3 setose setae (ss), longest 15-30 (24) µm long, one campaniform pore present only in some specimens; II: 3 ss, longest 32-38 (36) µm, campaniform pore (cp) present, 2-3 µm wide; III: 3 ss, longest 29-38 (33) µm long; IV: 1 fs, 9-15 (13) µm long; V: 1 ss, 22-28 (24) µm long and 1 fleshy seta (fs) 13 µm long; VI: 4 ss and 4-6 fs, longest apical seta 44-55 (48) µm long. Spiracles: all peritremes 12-13 µm wide; with 6 spiracular disc-pores, mesothoracic spiracle sometimes with 5-7 disc-pores, mainly quinquelocular, rarely with 3-4 loculi each about 3 µm wide, forming a band from each spiracle to body margin. Legs well developed, without tibio-tarsal sclerosis; claws with denticles; claw digitules unequal, one broad apically the other slender and knobbed at apex, each 16-19 (17) µm long. Coxae: 51-58 (54) µm long and 29-38 (33) μm wide; with 4 ss, longest 15-28 (20) μm long. Trochanter + femur: 74-83 (78) μm long, 22-25 (24) µm wide; trochanter with 2 ss, longest 19-35 (28) µm long; femur with 1 short ss. Tibia 51-57 (54) µm long and 15-16 (16) µm wide; with 2 ss, longest 13-17 (16) µm long. Tarsus 41-44 (42) µm long and 13-15 (13) µm wide; with 4 ss, longest 10-13 (11) µm long; tarsal digitules each 30-36 (33) µm long.

Figure 2. *Ceroplastes rusci* L., 2nd-instar male nymph.



Where, A = antenna and interantennal setae; B = spiracular disc-pores; C = ventral microduct with cruciform pore; D = metathoracic leg; E = abdominal ventral seta; Fi = ventral dermal spinules; Fii= dorsal dermal spinules; G = dorsal view of anal plates (left) and view of the ano-genital fold (right) ; H = view of the stigmatic cleft; I = dorsal tubular duct; J = marginal seta.

Comments: The 2^{nd} -instar male nymphs of *C. rusci, C. japonicus and C. sinensis* are very similar, the main difference appearing the number of spiracular disc-pores, marginal setae and dorsal tubular ducts as well. Spiracular disc-pores are fewer than in *C. japonicus*: they are 6 instead of 8-12 (10). This feature is shared with *C. sinensis*. Marginal setae are 8-9 between eyes, rather than 5-6 as in *C. japonicus*; 3-4 betweeen spiracular area, rather than 2 in *C. japonicus* and *C. sinensis*; marginal setae are 9-10 (9) between posterior spiracular area and anal cleft, in *C. rusci* and *C. sinensis* rather than 6 as in *C. japonicus*. Tubular ducts between eyespots are 11-13 in *C. rusci*, rather than 6-11 (9) as in *C. japonicus*. Claws differs from that of *C. japonicus* and *C. sinensis* by the presence of a small denticle. Bent marginal setae only occur rarely and only on head; a single longer marginal seta occurs on anal lobes rather than two, as in the other species.

PREPUPA (Fig. 3)

Described from 7 specimens in fairly good condition.

Unmounted material: brown reddish in colour.

Mounted material: body elongate, narrowest anteriorly, widest across abdomen, length 1073-1118 (1098 μ m), width across abdomen 521-611 (556 μ m). Division into head, thorax and abdomen not clear. Segmentation fairly defined, more distinct on abdomen.

Head: lacking mouthparts and simple eyes. Antennae fairly-well sclerotised, elongate, each 202-229 (218 μ m) long, with 9 poorly defined segments; antennal length to body length ratio 1:4.87-5.17 (5.05); antennal setae: IX: with 4-5 minute fleshy setae (*fs*).





Where: A = dorsal dermal spinules; B = ventral dermal spinules, and C = dorsal (left) and ventral (right) views of the posterior end of the abdomen. And where ab II-VIII = abdominal segments II-VIII; ads = dorsal abdominal setae; asp = anterior spiracle; avs = ventral abdominal setae; ce_{VII} = caudal extension of segment VII; ce_{VIII} = caudal extension of segment VII; h = head; ps = penial sheath; psp = posterior spiracle; sdp = spiracular disc-pore; vpls = ventropleural setae; vts = ventral thoracic setae; wb = wing bud

[...] With 2, sometimes 3, pair of minute interantennal setae ventrally, each 3-4 μ m long, and 2 minute setose dorsal head setae, each 4-9 (7) μ m long.

Thorax: prothoracic legs directed anteriorly, other pairs directed posteriorly; claws and digitules extremely reduced; metathoracic legs each 167-215 (191 μ m) long. Wing buds each 185-277 (236) μ m long, 85-96 (91) μ m wide, ratio of length to width 1: 0.34-0.46 (0.38). Spiracles: width of peritremes 17-20 (19) μ m, each with a group of 3 spiracular disc-pores near atrium; pores mainly with six loculi and about 4 μ m wide, but some number of loculi is highly variable. Setae: on each side, 1-2 minute thoracic setae near each coxa, 1 minute fleshy ventropleural seta on mesothorax.

Abdomen: segmentation fairly well defined; minute dermal spinules apparently most frequent posteriorly; dermal spinules with a different pattern on dorsum with respect to venter. Setae: pairs of minute dorsal abdominal setae present medially on segments V-VIII, each about 4-6 μ m long; with a pair of ventral abdominal setae, each 4-6 μ m long, on segments III-VIII. Margins with minute ventropleural setae, each 6-13 (9 μ m) long, segmentally arranged, with a single seta on each side of segments. Caudal extension VII (*ce_{VII}*) sclerotised, 46-58 (52) μ m long and 22-25 (24 μ m) wide, with 2 apical hair-like setae, each 26-32 (30) μ m long, and one shorter seta shortest 10-13 (11) μ m long; with also 1 dorsopleural seta at base of ce_{VII}. Caudal extension VIII (*ce_{VIII}*) poorly developed and fairly sclerotised. Penial sheath sclerotised, triangular in shape, 36-38 μ m long and 34-36 μ m wide, ratio of length to width is close to 1, with 2 minute penial sheath setae dorsally.

Comments: prepupa fairly larger than in *C. japonicus*. Any seta is present on antennal segment VIII and VII; in *C. japonicus* reduced setae are present on this segments. More head setae and interantennal setae occur on head with respect to *C. japonicus*. Only 3 spiracular disc pores are present in *C. rusci*, rather than 6-12 (8) as *in C. japonicus*. Any seta occurs on ce_{viii}.

PUPA (Fig. 4)

Described from 14 specimens in good condition.

Unmounted material: brown reddish in colour.

Mounted material: elongate oval, narrower anteriorly, widest across abdomen, length 967-1222 (1126) μ m, maximum width across abdomen 447-492 (473) μ m. Derm membranous. Head, thorax and abdomen not clearly divided. Segmentation not well defined, most distinct on abdomen.

Head: Derm membranous, sometimes showing fair sclerotisation, lacking mouthparts and simple eyes. Antennae fairly sclerotised, elongate, directed posteroriorly, each antenna with 9 poorly defined segments, 370-529 (477 μ m) long; antennal length to body length ratio 1:2.31-2.62 (2.34); with several minute fleshy setae towards the apex of terminal segments; antennal setae distribution: IX: 7; VIII: 1, VII: 1; each about 3 μ m long. With 2 pairs of dorsal head setae anteriorly on head, each about 9-10 μ m long; ventrally with 2 pairs of head setae, each 6-7 μ m long.

Thorax: legs well developed, each with clear segmentation; prothoracic legs are C – shaped and extend directed anteriorly, almost meeting in front of the head, other pairs directed posteriorly; procoxae with one minute seta; metathoracic legs each 463-555 (490) μ m long. Wing buds each 444-555 (491) μ m long and 130-185 (156) μ m wide; ratio of length to width 1:0.29-0.33 (0.31). Spiracles: width of peritremes 19-23 (22) μ m, each anterior spiracle with 8-11 (8) spiracular disc pores near atrium, each about 4 μ m wide, and each posterior spiracle with 3-5 (4) disc pores; number of loculi highly variable. Setae: ventrally with 1 very small thoracic seta near each coxa.

Abdomen: derm membranous; with rows of minute dermal spinules; dermal spinules with a different pattern on dorsum with respect to venter; segmentation fairly defined. Setae: with a pair of dorsal abdominal setae (*ads*), each about 4 μ m long, on segments V-VII; 2 ventral abdominal setae (*avs*) on segments III-VII, each about 4 μ m long + 2 ventropleural setae (*vpls*) on each side of segments IV-VII, each seta 9-10 μ m long; ante anal setae not detected. Caudal extensions on segment VII (ce_{VII}) well developed and fairly sclerotised, each 83-97 (93) μ m)long and 29-58 (44) μ m wide at base; each with 2 spinose setae, 25-38 (32) μ m and 22-33 (28) μ m long respectively, + 1 hair-like seta, 12-22 (16) μ m long. Caudal extensions on segment VIII (ce_{VIII}) poorly developed and fairly well sclerotised, each 18-29 (23) μ m long and 18-32 (24) μ m wide at base. Penial sheath (*ps*) elongate-triangular, sclerotised, 120-167 (148) μ m long, 73-96 (89) μ m wide at base, ratio of length to breadth 1:0.60 ; dorsally with 3 pairs of minute *hs*. Anal opening about μ m wide.

Discussion: Body dimensions agree with data provided by Silvestri & Martelli (1908), when they report total body length of 1,45 mm, body width of 0,58 mm, and wing buds length of 0,45 mm. The only noticeable difference from *C. japonicus* pupa consists in the number of spiracular disc-pores, that is lower than that reported for *C. japonicus*: anterior spiracle: 8-11 (8) spiracular disc-pores, rather than 10-16 (13); posterior spiracle: 3-5 (4) spiracular disc-pores, rather than 7-10 (9). Any seta occurring on ce_{viii} .

Figure 4. Ceroplastes rusci L., pupa.



Where A = ventral dermal spinules; B = dorsal dermal spinules. And where ads = dorsal abdominal setae; asp = anterior spiracle; avs = ventral abdominal setae; dpls = dorsal pleural setae; $ce_{VII} =$ caudal extension of segment VII; $ce_{VIII} =$ caudal extension of segment VIII; h = head; ps = penial sheath; psp = posterior spiracle; sdp = spiracular disc-pores; vpls = ventropleural setae; vts = ventral thoracic setae; wb = wing bud.

THIRD INSTAR NYMPH FEMALE (Fig. 5)

Described from 8 specimens in good condition. Details checked on remaining specimens.

Unmounted material: body oval, wider at abdomen, lightly convex dorsally, brown in colour.

Mounted material: body elongate oval; 789-954 (854) µm long; 536-954 (854) µm wide; anal cleft short.

Dorsum: Derm membranous. Small dorsal setae, rather short and bluntly spinose, each about 4-6 µm long, sparsely distributed throughout dorsum except clear areas, these more frequent on margin and head. Dorsal pores of two types. Dorsal pores mostly Ceroplastes-type (i), with a large central pore with predominantly one, occasionally two, satellite pores; pores abundant throughout dorsum, except on the lateral lobes or clear areas. Each central pore with long branched inner filament; pore opening heavily sclerotised, each 4-6 µm wide. Other pores (ii) present on dorsum with simple pore but with long inner filamentous, each with pore opening 3-4 µm wide. Anal plates each sub-triangular, with inner margins diverging, posterior margin convex with a rounded apex; dimensions of each plate: 118-144 (131) µm broad, anterior margin 144-163 (154) µm long, posterior margin 170–185 (176) µm long, interior margin 203–210 (206) µm long. Each plate with 1 apical seta 23-30 (25) µm long, 2 inner margin setae, the latter in a subapical position, setae 36-46 (41) µm and 22-39 (33) µm long respectively. Anogenital fold with 3-4 pairs of setae along anterior margin and 1 lateral margin seta. A couple of hypopygial setae on ventral surface just anterior to the ano-genital fold. Anal ring with 3 pairs of setae.

Figure 5. *Ceroplastes rusci* L., 3rd instar female.



Where, A = antenna; B = spiracular disc-pores (i) and B= view of stigmatic cleft (ii); C = ventral microduct with cruciform pore; D = metathoracic leg; E = marginal seta with shorther abdominal ventral seta; Fi = view of the ano-genital fold (i) and dorsal view of anal plates (ii); Gi= *Ceroplastes*-type dorsal pores (i) and view of pore opening (ii); H= dorsal setae.

Margin: Marginal setae setose, sometimes slightly bent, each seta 10-14 (13) μ m long; with 6-9 (8) setae between eyespots, 2-3 between eyespot and anterior stigmatic area; 2-3 laterally between stigmatic areas and 8-12 (10) between posterior stigmatic area and anal lobe; with a group of 3 longer setae on each side of anal cleft; longer seta 43-55 (50 μ m) long; shorter setae 23-40 (32) μ m long. Stigmatic cleft shallow or sometimes absent; stigmatic setae differentiated from the others, groups of conical spines along margin of cleft, central spine larger; with 7-11 (8) spines in anterior cleft and 5-8 (7) in posterior cleft; each spine 9-13 (12) μ m long and about 7 μ m wide at base. Width of each eyespot lens 23–26 (25) μ m.

Venter: derm membrabous. Dermal spinules present on mainly on abdomen and around genital opening. Ventral microducts quite heavily sclerotised, each with cruciform pore, sparsely distributed along margins, scarce throughout rest of venter. Ventral setae: with a single pair of moderately long pregenital setae; also two pairs of interantennal setae; longer seta 14-30 (21) µm long and shorter seta 60-89 (80) µm long; other very short ventral setae, each 2-4 µm long, distributed in fairly definite medial, sub- medial and sub-marginal lines on head and abdomen. Antennae 6 segmented, 152-178 (170) µm long, 3rd segment longest; dimensions: segment I: 17-26 (22) µm long and 32-36 (34) µm wide; II: 25-35 (32) µm long and 17-26 (22) µm wide; III: 55-70 (59) µm long and 17-22 (19) µm wide; IV: 14-17 (16) µm long and 15 μm wide; V: 16-17 μm long and 12-17 (13) μm wide; VI: 29-32 (31) μm long and 10-15 (12) µm wide; antennal setae: segment I: 3 setose setae (ss), longest 36-51 (43) µm long; II: 3 ss, longest 39-55 (49) µm, campaniform pore (cp) present, about 4 µm wide; III: 3 ss, longest 43-55 (51) µm long; IV: 1 fs, 16-22 (19) µm long; V: 1 ss, 16-28 (22) μ m long, and 1 fleshy seta (fs) 14 -19 (17) μ m long; VI: 4-6 ss and 3 fs, longest fs 22-30 (25) µm long, apical seta 30-55 (41) µm long. Spiracles: all peritremes19-20 (20)

μm wide; spiracular disc-pores, mainly with five loculi, rarely trilocular pores, forming a single pore band from each spiracle to body margin; each band becoming 2-3 pores broad along margin of clefts; anterior spiracular area with 7-9 (8) pores and posterior spiracular area with 10-11 pores; each pore 2-4 μm wide. Legs well developed, without tibio-tarsal sclerosis; claws with one small denticles; claw digitules unequal, one broad apically the other slender and knobbed at apex, each 22-30 (26) μm long. Coxae: 90-99 (93) μm long and 39-55 (47) μm wide; with 4 *ss*, longest 22-32 (27) μm long. Trochanter + femur: 96-113 (107) μm long, 28-35 (32) μm wide; trochanter with 1-3 short *ss*, longest 30-44 (36) μm long; femur with 1-2 short *ss* 12-19 (15) long. Tibia 70-78 (74) μm long and 19-20 (19) μm wide; with 2 *ss*, longest 17-20 (20) μm long. Tarsus 52-57 (54) μm long and 15-17 (15) μm wide; with 4 *ss*, longest 12-16 (15) μm long; tarsal digitules each 39-48 (42) μm long.

Discussion

Ceroplastes nymph instars are currently almost undescribed and 3rd-instar descriptions are only provided for *C. sinensis* (Qin & Gullan, 1994), *C. japonicus* (Camporese & Pellizzari, 1994) *C. destructor* (Wakari & Giliomee, 1998) and *C. pseudoceriferus* (Kawai & Tamaki, 1967). Differences mainly regard anal plates setae, that are 3 in *C rusci, C. pseudoceriferus* and *C. destructor* and 4 in *C. sinensis*; in *C. rusci* claws have a small denticle, that is absent in *C. sinensis* and *C pseudoceriferus*.
MALE MORPHOLOGY: CONCLUSIONS

Morphological affinities based on Ceroplastes adult males

Detailed descriptions of *Ceroplastes* adult males are available for very few species. In addition to *C. rusci* and *C. japonicus*, the other described adult males are *C. cirripediformis*, *C. ceriferus* (Gimpel *et al.*, 1974), *Waxiella berliniae*, and a *Waxiella* spp. (probably *C. mimosae* Signoret, according to Giliomee, 1967). Qin & Gullan (1995), at the end of their cladistic analysis of wax scales, concluded that "Cladistically, all wax scale should be included in one genus *Ceroplastes*"; moreover, with regard to male morphology of *C. berliniae* (= *W. berliniae*), *C. ceriferus*, *C. cirripediformis* and *C. sp.* (= *Waxiella* sp.), they stated that "The morphological differences among species are small, although they represent two currently recognized genera (*Ceroplastes* and *Waxiella*)". For the above reasons, the morphological characters of the described *Waxiella* males are here compared with those of *Ceroplastes*. The main differences appear to be:

1) Distribution of fleshy setae on penial sheath: the presence of fleshy setae on the basal portion of penial sheath is an unusual character that *C. japonicus* shares with *C. rusci* and *C. cirripediformis*, a Neotropical species. *C. ceriferus* does not have fleshy setae near the basal portion of penial sheath. *W. berliniae* and *Waxiella* sp., both African species, don't have any fleshy setae on penial sheath.

3) Occurrence of glandoular pouches: glandoular pouches on abdominal segment VIII occur on *C. rusci, C. cirripediformis* and *Waxiella* species, this feature is not shared with *C. japonicus* and *C. ceriferus,* a species considered by Qin *et al.* (1998) to be native to the Neotropical region.

3) Shape of penial sheath apex: the penial sheath apex is pointed in *C. rusci*, *C. japonicus*, *C. cirripediformis*, *W. berliniae* and *Waxiella* sp. The penial sheath apex is broadly rounded in *C. ceriferus*.

Other morphological affinities that may be here discussed concern the number of scutal setae that is nearly the same in the species studied, except *C. ceriferus*, where about 45 scutal setae occur. With regard to penial sheath length, the penial sheath is clearly longer on *C. rusci* (344-381 (361) μ m long). and *C. japonicus* (length: 244–310 (267) μ m) than on *C. cirripediformis* (length: 186 μ m). The examined morphological characters are resumed in Table 1.

On the basis of the examined characters, *C. rusci* and *C. cirripediformis* (which is believed to be a Neotropical species (Qin *et al.*, 1998)) share more diagnostic characters. *C. rusci* is thought to be native to the Afrotropical region (Qin et al., 1998, 1994;) or to the Mediterraean basin (Balachowsky, 1933). *C. ceriferus* does not show strong affinities with the other *Ceroplastes* adult males and has been separated from the other species, due to its peculiar penial sheath features. *C. japonicus*, has been considered to be an oriental species (Borchsenius, 1957) and is widely distributed in China, Korea and Japan, whereas in Abkhazia, Georgia and European countries it is clearly an alien species (Borchsenius, 1957; Japoshvili, 2001, *pers. com.*). However, Qin *et al.* (1998) considered it unlikely to be native to Asia and support a neotropical origin. Finally, *Waxiella* species do not share important morphological characters with other known *Ceroplastes* species and clearly form a well separated group.

Key to Ceroplastes species based on adult males

1 Penial sheath with fleshy setae	2
- Penial sheath without fleshy setaeGenus Waxiella	ı spp.
2 Penial sheath apex rounded; fleshy setae absent on the basal portion of penial sl	heath;
glandular pouches absentC. cere	iferus
- Penial sheath apex pointed; fleshy setae present on the basal portion of penial sl	heath;
glandular pouches present or absent	3

3 Glandular pouches absent	C. japonicus
- Glandular pouches present	4

4	Penial	sheath	long	(ratio	of	total	body	length	to	penial	sheath	length)
	1:3.7											C. rusci
-	Penial	sheath	shorter	(ratio	of	total	body	length	to	penial	sheath	length
	1:6.2			•••••						C	. cirriped	diformis

Table 1. Comparison of some morphological characters and range of males of *Ceroplastes* and *Waxiella* species. Morphological data for*C. cirripediformis* and *C. ceriferus* taken from Gimpel *et al.* (1974); data for *W. berliniae* and *Waxiella* spp. taken from Giliomee (1967).

Species	Distribution of penial sheath fleshy setae	Glandular pouches and associated setae	Shape of penial sheath apex	Number of scutal setae <i>fs</i>	Number of scutal setae <i>hs</i>	Ratio of total body-length to penial sheath length	Supposed origin
C. rusci	Absent on apical portion of penial sheath	Present	Pointed	10-12 (11)	6-10 (8)	1: 3.7	Afrotropical or Mediterranean Oriental
C. japonicus	Absent on apical portion of penial sheath	Absent	Pointed	10-14 (12)	6-10 (8)	1: 4.0	or Neotropical
C. cirripediformis	Absent on apical portion of penial sheath	Present	Pointed	fs + hs:	about 23	1: 6.2	Neotropical
C. ceriferus	Absent on the basal portion of penial sheath	Absent	Rounded	fs + hs:	about 45	1: 4.9-5.2	Neotropical
W. berliniae	Absent	Present	Pointed	10-20 (14)	4-14 (10)	1: 3.6 - 4.1 (3.8)	Afrotropical
<i>Waxiella</i> spp.	Absent	Present	Pointed	2-11 (6.7)	9-12 (11)	1: 3.5 - 3.8 (3.6)	Afrotropical

SECTION III

REDESCRIPTION OF THE ADULT FEMALE AND FIRST-INSTAR NYMPH OF *CEROPLASTODES DUGESII* (SIGNORET, 1886) (HEMIPTERA: COCCIDAE) AND DESCRIPTION OF THE OTHER IMMATURE STAGES

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I contributed to all the parts of the work and to the paper writing

Introduction

Lecanopsis dugesii was briefly described by Signoret (1886) on the basis of specimens sent to him by Lichtenstein. The genus Ceroplastodes was introduced by Cockerell (1893b) to take his species *Inglisia nivea* (Cockerell, 1893a). Later, in 1902, Cockerell synonymised C. nivea (Cockerell) with L. dugesii Signoret. Recently, Avasthi and Shafee (1991) studied the types of C. dugesii, C. daleae Cockerell and C. deani Lawson preserved at U.S. National Museum of Natural History, Washington (USNM), and synonymized C. daleae Cockerell and C. deani Lawson with C. dugesii; they also gave a short redescription of the adult female. Hodgson (1994) redescribed the adult female on the basis of the type of C. dugesii preserved at USNM. Because of the poor condition of the specimens (old post-reproductive females), neither Avasthi and Shafee (1991) nor Hodgson (1994) were able to fully describe the dorsum. More recently Hodges and Williams (2003) described the 1st-instar nymph, also from specimens preserved at the USNM. It is clear from the data labels reported in the cited papers (Avasthi & Shafee, 1991; Hodgson, 1994; Hodges & Williams, 2003) that the recent descriptions and redescriptions of C. dugesii were based on the same material, collected between 1894 and 1906 and preserved at the USNM. Because most of the life stages of this species, the type species of the genus, were recently collected off Acacia sp. in Hidalgo and Coahuila States (Mexico), the opportunity is here taken to redescribe the adult female and the 1st-instar nymph and to describe the remaining immature stages. C. dugesii is currently only known from Mexico and the USA (Kansas and New Mexico) on Mimosaceace and Fabaceae (Acacia sp., Mimosa sp., Dalea formosa, Kuhnistera purpurea, Petalostemon violaceum and Prosopis pubescens).



Plate 1. Tests of *C. dugesii* (Signoret) on *Acacia* sp. Above: the larger, white, waxy, glassy tests of the adult females mixed with the smaller white male tests. Below: tests of adult females of C. dugesii and some smaller, brown, flat immature stages (photos P. Fontana).



Plate 2. Branch of Acacia sp. infested by C. dugesii (Signoret) (photos P. Fontana).

Material studied

The material studied was collected from two sites in Mexico off Acacia sp., namely: State of Hidalgo, 23rd September, 2004, along the road to Queretaro, at 2265m ASL; and State of Coahuila, 1st July, 2005, along road from Arteaga to Matehuala, at 2094m ASL. Terminology follows that of Hodgson (1994). The stigmatic area is here referred as the area between the two outermost spiracular disc-pores of the margin.

Specimen depositories

Slides of the mounted and unmounted specimens of all available stages are deposited in the collection of the Department of Environmental Agronomy and Crop Production–Entomology (DEAE), University of Padua, Italy (slides numbers: 1191; 1194). Specimens are also deposited in BMNH.

Ceroplastodes dugesii (Signoret)

Lecanopsis dugesii Signoret, 1886: XXXIX.

Inglisia nivea Cockerell, 1893a: 160. Nomen nudum.

Fairmairia (Ceroplastodes) nivea Cockerell; Cockerell, 1893b: 350.

Ceroplastodes daleae Cockerell, 1894:13.

Ceroplastodes dugesii (Lichtenstein and Signoret): Fernald, 1903: 164; Avasthi and Shafee, 1991:1.

Ceroplastodes deani Lawson, 1917: 203.

Ceroplastodes dugesii (Signoret); Ben-Dov, 1993: 61; Hodgson, 1994: 167; Williams and Hodges, 1997: 144; Hodges and Williams, 2003: 496.

Material studied: Mexico, Hidalgo, road to Querétaro, 2265m ASL, 23.ix.2004, P. Fontana (DEAE, No. 1191): 8 adult females, 4 3rd-instar females, prepupa and pupa (DEAE: No. 1191); also No. 1191: 2 adult females, 3 3rd-instar females, 1 2nd-instar female, 2 2nd-instar males, 6 1st-instar nymphs (BMNH); Mexico, Cohauila, 1.vii.2005, P. Fontana (DEAE, No. 1194): 16 3rd-instar females, 4 2nd-instar females, 18 1st-instar nymphs (DEAE).

ADULT FEMALE (Fig. 1)

Unmounted material.Living specimens covered with a white, waxy, glassy test; oval, convex, with a rough surface and small protuberances (Plates 1 and 2).

Mounted material. (Described from 8 specimens in good condition): oval, dorsum wider than venter; length 2235–4470 (3189) m, width 1565–2980 (2332) m.

Dorsum. Membranous, wider than venter. Preopercular pores, each round to oval, of varying size (largest about 7 µm wide); in two sparse diverging groups, each with 17– 33 (23) pores, approximately on abdominal segments III-V anterolaterally to anal plates. Other dorsal pores of 3 or 4 types present, all tending to be frequent to abundant medially and marginally, perhaps less frequent submarginally: (i) minute microductules, each about 0.5–1.0 µm wide with a long narrow inner ductule about 17 µm long: frequent; (ii) a slightly larger pore about 1.5 µm wide, also dark but apparently without an inner ductule, frequent throughout; (iii) a simple pore about 2 µm wide, without a central spot: frequent throughout and in a submarginal line; and (iv) a larger pore about 3 µm wide, with a central darker area, rather resembling a setal socket: scattered throughout but apparently in two diverging lines on abdomen anterior to anal plates. Anal plates each sub-triangular, with inner margins diverging; dimensions of each plate (m): 80–116 (100) broad, anterior margin 101–131 (112) long, posterior margin 87–131 (112) long, inner margin 101–131 (123) long; each with 2 conical spinose setae along inner margin, each seta 32-41 (35) µm long, a single apical seta 29–38 (33) µm long, and 2 setae on outer margin, 43–68 (48) µm and 58– 87 (74) µm long respectively; more apical setae similar to others, but more anterior setae long and curved. Anogenital fold with 2 pairs of setae along anterior margin and 1 long subapical seta on each lateral margin. Anal ring broad, with many pores and 8 pairs of setae.

Margin. Marginal setae spinose, conical, distributed along body margin, each seta 11– 37 μ m long and about 6 μ m wide at base; with 40–78 (60) anteriorly between eyespots; 25–40 (34) between eyespot and anterior stigmatic area; 32–51 (42) laterally between stigmatic areas, and 101–121 (109) between posterior stigmatic area and anal lobe; with a spinose seta along each anal cleft margin. Stigmatic cleft absent; stigmatic setae similar to marginal setae but slightly larger, usually displaced on a double row along margin; with 5–11 (8) setae in each anterior and 4–11 (7) in each posterior stigmatic area; each seta 16–32 (23) μ m long and 6–9 (7) μ m wide at base. Width of each eyespot lens 22–25 (23) m.

Venter. Derm membranous. Dermal spinules present on thorax and abdomen. Pregenital multilocular disc-pores each 6–9 (7) um wide and mainly with 7–11 loculi, present medially and mediolaterally on all abdomial segments and on metathorax. Spiracular disc-pores, each 4–5 µm wide and with mainly 5 (range 2–8) loculi, present in a band from each spiracle to body margin, each band broadening near margin; with 33–43 (36) in each anterior band and 27–58 (38) pores in each posterior band. Ventral microducts sparse, present more or less throughout but absent medially on abdomen. Ventral tubular ducts of two types: (i) a duct with fairly broad outer ductule (each 17– 28 (22) µm long, 4–6 µm wide), a thin inner ductule (10–15 (12) µm long), and with a well-developed terminal gland; in a broad marginal band, but becoming infrequent on either side of anal cleft; band broadens considerably between antennae; similar ducts also present sparsely medially on head, thoracic and abdominal segments; and (ii) a duct with outer ductule about twice width of type (i), each 13–16 µm long, 5–7 µm wide, with filamentous inner ductule without a terminal gland; restricted to either side of anal cleft. Ventral setae mainly short, each 7–12 (9) μ m long, distributed in fairly definite medial, sub-medial and sub-marginal lines. With four pairs of interantennal setae, of which three pairs short (each 7–9 µm long) and one pair distinctly longer (each 9–17 (12) μ m long); with two small setae on head, near apex, 6–10 (9) μ m long; also a pair of longer pregenital setae, each 20-38 (32) µm long. Antennae 7 segmented, each 265–330 (289) µm long; 4th segment longest, usually with a distinct pseudo-segmentation, causing antenna to appear 8 segmented. Preantennal pore present, 3-4 µm wide. Clypeolabral shield 185-195 µm long; labium with 4 pairs of setae. Spiracles each rather large, width of peritremes: anterior 50-60 m, posterior 55-65 m; each with a sclerotised spiracular plate. Legs well developed, lengths (m): trochanter + femur 206–236 (218); tibia + tarsus 254–283 (271); tarsal digitules 38–41 (39); claw 22–27 (24); claw digitules 19–22 (21), broad apically; tibia and tarsus

separate, without an articulatory sclerosis; claw with denticle. Vulva present between abdominal segments VII and VIII.

Comments: the distribution of the dorsal pores was hard to discern. The larger "simple" pore (on all stages apart from the 1st-instar nymph) closely resembled a setal socket, and many had a dark spot centrally. Similar pores have been noted on some Australian species (unpublished). Avasthi and Shafee (1991), in their redescription of the adult female of *C. dugesii*, recognized "numerous oval pale areas each with a minute pore" on the dorsum, and these probably refer to the preopercular pores. On two of the specimens, two tubular ducts were also noted on the dorsum, approximately submedially on abdominal segment IV, but these are thought to be an artefact.



Figure 1. Ceroplastodes dugesii (Signoret), adult female.

Where, Ai and Aii = ventral tubular ducts; B = preopercular pore; C = pregenital discpore; D = spiracular disc-pore; E = dorsal simple pore; F = ventral microduct; G =dorsal microductule; H = marginal seta; I = ventral seta; J = dorsal pore resemblingsetal socket; K = antenna; L = metathoracic tarsus and claw; MI = anal plates, dorsalview, MII = anal plates, ventral view, and N = spiracle.

FIRST-INSTAR NYMPH (Fig. 2)

Mounted material. (Described from 18 specimens): oval, length 413–525 (471) m, width 200–295 μ m (259).

Dorsum. Derm membranous, anal lobes well defined. Simple pores, each 1.0–1.5 μ m wide, in 2 submedial and 2 submarginal lines, with perhaps 11 pores in each median line and 14 in each submarginal line. Microductules minute, each usually close to a simple pore and tending to be located in or close to an intersegmental membrane. Anal plates each sub-triangular, with a concave inner margin; dimensions of each plate (m): 29–33 (30) broad, anterior margin 26–39 (35) long, posterior margin 25–32 (29) long, inner margin 43–55 (50) long; each with 2 setae along inner margin, 15–42 (23) μ m and 25–29 (27) μ m long respectively, a single apical seta 124–181 (153) μ m long, and a single seta on outer margin 14–16 μ m long. Anal ring with 6 setae.

Margin. Marginal setae spinose and conical, each 6–9 (8) μ m long and 4–7 (5) μ m wide at base, distributed along body margin as follows: 6 between eyespots and (on each side) 2 between eyespot and anterior stigmatic area; 2 or 3 between stigmatic areas; 8 between posterior stigmatic area and anal lobe. Stigmatic cleft absent. Spiracular setae undifferentiated from marginal setae, usually with 2 setae associated with each furrow, often with one slightly displaced onto dorsum. Eyes present on margin; width of each eyespot lens 7–10 (8) m.



Figure 2. Ceroplastodes dugesii (Signoret), 1st-instar nymph.

Where, A =spiracular disc-pore; B =dorsal simple pore; C =ventral trilocular pore; D =dorsal microductule; E =ventral microduct; F =marginal seta; G I =and G II =ventral setae; H =antenna, and I =metathoracic leg.

Venter. Membranous, with well-defined segmentation. Dermal spinules fairly defined on abdomen. A pair of sclerotised trilocular pores present near apex of head anterior to each scape, each $3-4 \mu m$ broad. With one spiracular disc-pore, $4 \mu m$ wide, usually with 5 (rarely 4 or 6) loculi, located between spiracles and body margin. Ventral microducts: 1 or 2, each 1.5 µm wide, present on mesothorax posterior to each anterior spiracle. Very small ventral setae, each 1–2 µm long, distributed in medial, inner submarginal and sub-marginal rows on all abdominal segments; medial setae on segments VI and VII each 14–25 (19) µm long; with 1 pair of interantennal setae, each 20–26 (23) μ m long; with two setae on head, near margin, each 7–17 (13) μ m long. Antennae six segmented, 141-171 (157) µm long, 3rd and apical segments longest; setal distribution showing nothing distinctive. Preantennal pore not detected. Clypeolabral shield 58-87 (68) µm long; labium with 3 or 4 pairs of setae. Width of spiracular peritremes: anterior 7-9 m, posterior 9 m; sclerotised spiracular plates present. Legs well developed, without a tibio-tarsal sclerosis and without microspines along distal margin; claws 18-30 (22) µm long, with a denticle; claw digitules each with small apices and 23–28 (25) µm long; tarsal digitules longer than claws, offset, one longer than other, longest 30-39 (33) μ m long; tibia + tarsus 148–195 (170) μ m long; trochanter + femur 89–118 (101) µm long.

Comments: The above description agrees with that of Hodges and Williams (2003), but with an important difference: the trilocular pores appear to be clearly in a ventral position, whereas on the other known 1st-instar nymphs of soft scales they are usually found on the dorsum. These pores were more conical and more heavily sclerotised than those normally found dorsally. In addition, on our specimens, the marginal spinose setae are more conical and the tarsal and claw digitules are longer than in their fig.1. Hodges and Williams also state in the text that two spiracular disc-pores are present in each spiracular furrow but only one is illustrated in their Fig.1 (as on our specimens). On the other hand, the same authors, in their paper on taxonomic characters of nymphs (Williams & Hodges, 1997), present some details of *C. dugesii* 1st-instar nymph (marginal setae, spiracular setae, leg) that are similar to our specimens.

SECOND-INSTAR FEMALE (Fig. 3)

Unmounted material. Body oval, lightly convex dorsally, derm membranous, brown in colour.

Mounted material. (Described from 4 specimens in good condition): oval, length 805–969 (843) m, width 358–536 (471) m. Anal cleft short, apex of anal plates approximately level with anal lobes.

Dorsum. Pores of 3 types present: (i) simple pores, each about 1.5 μ m wide, scattered over dorsum and in a submarginal band; (ii) microductules, each less than 1 μ m wide, in a sparse marginal band and probably frequent elsewhere; and (iii) a larger pore, each about 2.5–3.0 μ m wide with a sclerotised outer margin, in two diverging longitudinal lines anterior to anal plates, but rather scattered; generally with 1 pair on each abdominal segment and then perhaps more frequent on thorax and head. Anal plates each sub-triangular, broad in shape, with inner margins diverging; dimensions of each plate (m): 36–87 (58) broad, anterior margin 36–87 (55) long, posterior margin 51–109 (78) long, inner margin 70–94 (78) long; each with 2 spinose setae along inner margin, 12–19 (16) μ m and 17–20 (19) μ m long respectively, an apical seta 28–38 (31) μ m long, and a single seta on outer margin, 22 μ m long. Anogenital fold with 2 pairs of setae along anterior margin and 1 seta on each lateral margin. Anal ring with 6 setae.

Margin. Marginal setae spinose, conical, distributed along body margin, each seta 12– 17 (14) μ m long, 4–6 (5) μ m wide at base. Distribution of marginal setae: 14–16 (15) between eyespots; 5–8 (7) between eyespot and anterior stigmatic area; 6–8 (7) between stigmatic areas; 18–26 (22) between posterior stigmatic area and anal lobe; with 1 spinose seta along margin of anal cleft. Stigmatic cleft absent; stigmatic setae not clearly differentiated from marginal setae, numbering 3–4 in the anterior stigmatic area and 3 in the posterior stigmatic area. Measurements of stigmatic setae: 10–20 (15) μ m long and 4–7 (5) μ m wide in each anterior stigmatic area, 12–18 (14) μ m long and 4–7 (6) μ m wide in each posterior stigmatic area. Width of each eyespot lens 12–16 (14) m.

Venter. Membranous, with well-defined segmentation. Dermal spinules present on thorax and abdomen. Spiracular disc-pores, each 4 µm wide with mainly 5 loculi, forming a narrow band broadening from each spiracle to body margin, with 6–11 (9) pores in each anterior band and 6-9 (7) in each posterior band. Preantennal pores present, each 2–3 µm wide. Ventral microducts each 1.5 wide, with 1 or 2 on each side of thorax. Ventral tubular ducts with outer ductule 12–13 µm long, 3–6 (4) µm wide, inner ductule thin, 9–12 (10) µm long, and terminal gland 4 µm wide, forming a sparse submarginal band extending from anal lobes to head. With 3-8 (5) ducts between evespots, 2 or 3 between evespot and anterior stigmatic cleft; 2-5 (4) between stigmatic clefts, and 10–14 (12) between posterior stigmatic cleft and anal lobe. Small ventral setae on abdominal segments in well-defined medial, submedial and submarginal lines; medial setae on segment VII longer than others, each 17–29 (22) μ m long. Length of ventral setae: medial setae 3–4 μ m long, sub-medial setae 3–6 μ m long, sub-marginal setae 2-7 µm long. Other small setae present near coxae and on head; with 2 small setae near apex of head, each 4-7 (6) µm long; with 2 pairs of interantennal setae, one pair longer than other. Antennae six segmented, each 129–141 (135) µm long, segment III longest, sometimes with a weak pseudosegmentation in middle. Clypeolabral shield 73–102 (87) µm long; labium with 4 pairs of setae. Width of spiracular peritremes: anterior 13 m, posterior 12-15 m; each with a sclerotised spiracular plate. Legs well developed, without a tibio-tarsal sclerosis; claws with a denticle, each 15–19 (16) µm long; claw digitules both broad distally, each 16–19 (17) μm long; tarsal digitules of slightly different thicknesses, each 26–35 (30) μm long; tibia + tarsus 141–155 (151) μ m long; trochanter + femur 92–107 (98) μ m long.





Where, A = ventral tubular duct; B = spiracular disc-pore; C = dorsal simple pore; D = dorsal pore resembling setal socket; E = marginal seta; F = interantennal setae; G = antenna; H = metathoracic leg; I = dermal spinules; J = ventral microduct; K = dorsal microductule, and L = spiracle.

THIRD-INSTAR FEMALE (Fig. 4)

Unmounted material. Body oval, lightly convex dorsally, derm membranous, brown in colour.

Mounted material. (Described from 17 specimens in fair to good condition): oval, length 969–1430 (1167) m, width 626–864 (731) m. Anal cleft about twice length of anal plates.

Dorsum. Pores of 3 types present although distribution unclear: (i) simple pores, each about 1.5 μ m wide, scattered over dorsum and in a marginal/submarginal band; (ii) microductules, each less than 1 μ m wide, in a sparse marginal band and probably frequent elsewhere; and (iii) a larger pore, each about 2.5–3.0 μ m wide with a sclerotised outer margin, in two diverging longitudinal lines anterior to anal plates, but possibly only present on posterior 3 or 4 abdominal segments. Anal plates each subtriangular, broad, with inner margins diverging; dimensions of each plate (m): 43–65 (52) wide, anterior margin 43–65 (51) long, posterior margin 65–80 (70) long, inner margin 78–94 (86) long; each with 2 spinose setae along inner margin, 22–25 (25) μ m and 10–23 (16) μ m long, an apical seta 22–26 (24) μ m long, and 1 seta along outer margin 22–62 (45) μ m long. Anogenital fold with 2 pairs of setae on anterior margin plus one long seta, 36–39 (37) μ m long, on each lateral margin near apex of anal plates. Anal ring with 6 setae.

Margin. Marginal setae spinose and conical, distributed along body margin, each 14– 17 (16) μ m long, 6–7 μ m wide at base. Distribution of marginal setae: 29–34 (31) between eyespots; 12–16 (14) between eyespot and anterior stigmatic area; 13–19 (16) between stigmatic areas; 39–55 (48) between posterior stigmatic area and anal lobe; with 1, rarely 2, spinose setae along margin of anal cleft. Stigmatic cleft absent; stigmatic setae not clearly differentiated from marginal setae but generally slightly longer, forming a fairly defined group of 2 rows, numbering 6–7 in each anterior stigmatic area and 3–6 (4) in each posterior stigmatic area. Measurements of stigmatic setae: 14–20 (18) μ m long and 6–7 μ m wide in each anterior stigmatic area, 14–19 (16) μ m long and 6 μ m wide in each posterior stigmatic area. Width of each eyespot lens 17–22 (19) μ m.

Figure 4. Ceroplastodes dugesii (Signoret), 3rd-instar female nymph.



Where, A = ventral tubular duct; B = spiracular disc-pore; C = dorsal simple pore; D = preantennal pore; E = dorsal microductule; F = ventral microduct; G = dorsal pore resembling setal socket; H = marginal seta; I = submarginal seta on head; J = ventral abdominal seta; K = antenna; L = metathoracic leg; M = dermal spinules, and N = spiracle.

Venter. Membranous, with well-defined segmentation. Dermal spinules present on abdomen plus a few also on head and thorax. Spiracular disc-pores each 4 µm wide with mainly 5 loculi (rarely with 3, 4 or 6 loculi), forming a narrow band broadening from each spiracle to body margin; with 11-24 (17) pores in each anterior band and 14–23 (18) in each posterior band. Preantennal pores each 2 µm wide. Ventral tubular ducts of one type, each with outer ductule 13–15 (13) µm long and 3–4 µm wide, inner ductule 7-10 (9) µm long, with a well-developed terminal gland, 3-4 µm wide; forming an irregular, sparse submarginal band extending from anal lobes to head; present also medially near labium on head and on thoracic segments. Microducts, each 1.5 µm wide, in a sparse submarginal band. Small ventral setae (each 4–5 µm long) distributed in welldefined medial, sub-medial and sub-marginal lines on abdominal segments; medial setae on segment VII longest, each 23-41 (33) µm long. Other setae on thorax and head short and sparse; with 2 setae near apex of head, each 4–9 (7) µm long; usually with 3 pairs of interantennal setae, one pair longer than others, longest 7– 12 (10) µm long. Antennae 6 segmented, 177–307 (201) µm long; segment III longest, sometimes with a weak pseudosegmentation in middle. Clypeolabral shield 87–123 (104) µm long; labium with 4 pairs of setae. Width of spiracular peritremes: anterior 17–22 (20) m, posterior 20–26 (22) m; each with a sclerotised spiracular plate. Legs well developed, without a tibio-tarsal sclerosis; claws each 15–22 (19) µm long, with a denticle; claw digitules similar and broad distally, each 16-25 (19) m; tarsal digitules also alike, each 32–38 (34) µm long; tibia + tarsus 107–207 (187) µm long; trochanter + femur 118–148 (138) μm long.

SECOND-INSTAR MALE (Fig. 5)

Unmounted material. Body elongate oval, dorsally slightly convex, derm membranous, brown in colour.

Mounted material. (Described from 13 specimens, 4 in good condition): elongate oval, length 849–1699 (1292) m, width 447–1281 (745) m. Anal cleft short, apex of anal plates almost extending to level with anal lobes.

Dorsum. Pores of 3 types present: (i) simple pores, each about 1.5 µm wide, scattered over dorsum and in a submarginal band; (ii) microductules, each less than 1 µm wide, in a sparse marginal band and probably frequent elsewhere; and (iii) a larger pore, each about 2.5-3.0 µm wide with a sclerotised outer margin, in two diverging longitudinal lines of about 10 pores anterior to anal plates. Dorsal tubular ducts restricted to two groups mediolaterally on abdominal segment IV, each group with 20-28 (22) tubular ducts, each duct with an outer ductule 14–17 (16) μ m long and 4 μ m wide, a thin inner ductule 6–10 (8) µm long and a large terminal gland 3–4 µm wide; outer ductule with a cup-shaped invagination broader than on ventral tubular ducts. Anal plates each sub-triangular, broad, with inner margins diverging; dimensions of each plate (m): 29–58 (44) wide, anterior margin 33–58 (41) long, posterior margin 43–53 (57) long, inner margin 51–73 (64) long; each with 2 spinose setae along inner margin, 18–20 µm and 10–18 (15) µm long respectively, 1 long and bent apical seta, 29-36 (33) µm long, and a single seta on outer margin, 22-33 (28) µm long. Anogenital fold with 2 pairs of setae on anterior margin and with one long seta on each lateral margin. Anal ring with 6 setae.

Margin. Marginal setae spinose and conical, each 13–16 (15) μ m long, 6–7 μ m wide at base. Distribution of marginal setae: 17–23 (21) between eyespots; 8–11 (10) between eyespot and anterior stigmatic area; 10–12 (11) between stigmatic areas, and 29–36 (32) between posterior stigmatic area and anal cleft; with 1 spinose seta along margin of anal cleft. Stigmatic clefts absent; stigmatic setae not clearly differentiated from marginal setae, but generally appearing slightly longer, and with one slightly displaced onto dorsum, with 4–6 (5) in each anterior stigmatic area and 3–4 in each posterior

stigmatic area; each 13–16 (15) μ m long and 4–6 μ m wide in each anterior stigmatic area and 15–16 (15) μ m long and 6–7 μ m wide at base in each posterior stigmatic area. Width of each eyespot lens 15–17 m.

Venter. Derm membranous with well-defined segmentation. Dermal spinules present on abdomen. Spiracular disc-pores, each $3-4 \mu m$ wide and with mainly 5 (rarely 4) loculi, forming a narrow band between each spiracle to body margin, each band broadening near margin, with 8–16 (13) pores in each anterior band and 8–15 (10) in each posterior band. Microducts, each 1.5 µm wide, in a sparse submarginal band. Preantennal pores present, each 2–4 µm wide. Tubular ducts, each with a narrow inner ductule and large terminal gland similar to those on dorsum, distributed in a submarginal row extending from head to anal lobes and medially onto thorax and abdomen. Ventral setae all small, each 3-7 (5) µm long, distributed in well-defined medial, submedial and submarginal longitudinal lines on abdomen; medial setae on segment VII longest, each 17–30 (26) µm long; with two small setae on apex of head, each 7–9 µm long; with two pairs of interantennal setae, each 9–17 (13) µm long. Antennae 7 segmented, 142–177 (164) µm long. Clypeolabral shield 73–116 (87) µm long; labium with 4 pairs of setae. Width of spiracular peritremes: anterior 15–17 m, posterior 15–19 (17) m; each with a sclerotised spiracular plate. Legs well developed, without a tibio-tarsal sclerosis; claws each 15–22 (19) µm long, with a denticle; claw digitules similar and broad distally, each 14–17 (16) µm long; tarsal digitules both alike, each 27–30 (29) μ m long; tibia + tarsus 142–189 (171) μ m long; trochanter + femur 100–153 (115) µm long.

Comments. The distribution of the dorsal and ventral tubular ducts in the 2nd-instar male of C. dugesii resembles that on the 2nd-instar male of *Paracardiococcus actinodaphnis* Takahashi (Hodgson, 1994), a species also belonging to the *Cardiococcinae*.



Figure 5. Ceroplastodes dugesii (Signoret), 2nd-instar male nymph.

Where, A = ventral tubular duct; B = dorsal tubular duct; C = dorsal simple pore; D = preantennal pore; E = dorsal microductule; F = ventral microduct; G = spiracular discpore; H = dorsal pore resembling setal socket; I = marginal seta; J = interantennal setae; K = ventral abdominal seta; L = dermal spinules; M = antenna; N = metathoracic leg, and O = spiracle.

PREPUPA (Fig. 6)

Mounted material. (Described from 2 specimens): body elongate, narrowest anteriorly, widest across abdomen, length 1535–1565 m, width across abdomen 700–745 m. Derm membranous. Division into head, thorax and abdomen not clear. Segmentation indistinct, most defined on abdomen.

Head. Lacking mouthparts and simple eyes. Antennae fairly-well sclerotised, elongate, each 283–313 μ m long, with 9 poorly defined segments; antennal length to body length ratio 1:5.2. Setae: dorsally with a pair of minute fleshy setae, each 8 μ m long; ventrally with minute fleshy setae scattered on anterior of head, each 6 μ m long.

Thorax. Prothoracic legs directed anteriorly, other pairs directed posteriorly; claws and digitules absent; metathoracic legs each 242–372 μ m long. Wing buds each 313–336 μ m long, 147–177 μ m wide, ratio of length to width 1:0.5. Anterior spiracles: width of peritreme 22 m, each with 10 spiracular disc pores near atrium; posterior spiracles each with 1–4 disc pores; number of loculi in each spiracular pore highly variable. Setae: with two minute ventral thoracic setae, each 4 μ m long, just posterior to each procoxa, plus a pair of minute apparently fleshy ventropleural setae laterad to each procoxa, each seta 9 μ m long.

Abdomen. Segmentation fairly well defined. Setae: segments IV–VII with a pair of minute dorsal abdominal setae, each 3 μ m long, in a medial row; with 1 pair (occasionally 2 pairs) of ventral abdominal setae, each 6 μ m long, on segments II–VII. Margins with minute dorso-pleural setae segmentally arranged, with a single seta on





Where, ab II–VIII = abdominal segment II–VIII; ads = dorsal abdominal setae; apl = abdominal pleural setae; asp = anterior spiracle; avs = ventral abdominal setae; h = head; ps = penial sheath; psp = posterior spiracle; sdp = spiracular disc pores; vts = ventral thoracic setae; wb = wingbud.

[...] each side of segments I–IV and 2–3 fleshy setae on segments V–VIII. Segment II with a minute ventral abdominal seta, 3 μ m long, laterad to each metacoxa. Caudal extensions and anal opening absent. Penial sheath sclerotised, triangular in shape, wider than long (94–118 μ m long, 130–177 μ m wide, ratio of length to width 1:1.44), without minute pores on dorsal surface.

PUPA (Fig. 7)

Mounted material. (Described from 3 specimens, 2 in fair to good condition): elongate oval, narrower anteriorly, widest across abdomen, length 1565–1892 (1674) m, maximum width across abdomen 670–745 (710) m. Derm membranous. Division into head, thorax and abdomen unclear. Segmentation obscure, most defined on abdomen. Spiracular disc-pores present near anterior spiracular atrium. Tubular ducts and pores, except spiracular disc-pores, absent and setae few.

Head. Lacking mouthparts and simple eyes. Antennae elongate, fairly-well sclerotized, 10-segmented, directed postero-laterally, each 745–775 (755) μ m long; antennal length to body length ratio 1:2.22, without setae. Thorax. Legs well developed, segmentation clear; prothoracic legs directed anteriorly, with tibia and tarsus curved towards midline of body; other pairs directed posteriorly; metathoracic legs each 625–755 (700) μ m long. Wing buds extending posteriorly as far as anterior abdominal segments; each 590–667 (629) μ m long, 177–230 (210) μ m wide; ratio of length to width 1:0.33. Spiracles: width of each anterior peritreme 37 μ m wide,





Where asp = anterior spiracle; avs = ventral abdominal setae; ceVIII = caudal extension of segment VIII; dpls = dorsopleural setae; h = head; ps = penial sheath; psp = posterior spiracle; vpl = ventropleural setae; wb = wingbud.

[...] with 18–22 disc-pores near each atrium; number of loculi in each disc-pore highly variable (from 1 to 12); posterior peritreme not measurable on the available specimens.

Abdomen. Segmentation fairly well defined. Setae: 2 (rarely 1 or 3) minute dorsopleural setae, each 6–13 (10) μ m long, segmentally arranged on sides of segments III– VII; also with 1 pair of minute ventral abdominal setae, each 9–13 (10) μ m long, medially on segments IV–VII; also with pairs of ventropleural setae on segments VI and VII; ante-anal setae absent. Caudal extensions on segment VIII very reduced, each 16–51 (28) μ m long and 26–58 (44) μ m wide at base; each with 2 minute fleshy setae and 1 minute hair seta 4–7 (6) μ m long. Penial sheath triangular, sclerotised, 153–177 (163) μ m long, 136–148 (142) μ m wide at base, ratio of length to breadth 1:0.87; with some minute pores, each about 1 μ m wide, on dorsal surface. Anal opening not seen.

Discussion

The 1st-instar nymph, 2nd-instar male and female nymphs, 3rd-instar nymph and the adult female of *C. dugesii* are characterised by the constant presence of 2 ventral setae near the apical margin of the head: these setae are long and well developed in the 1st-instar nymph, small but clearly discernible on the older nymphs and adult female. The ventral derm of the nymphs and adult female is membranous, but with distinct segmentation visible on the thorax and abdomen. Two distinctive features of *C. dugesii* are the presence of two diverging bands of preopercular pores on the abdominal region of dorsum on the adult female, and the presence on the first instar nymph of the trilocular pores in a ventral position, instead of in a dorsal position, as is usual on first instar.

CHAPTER II

ECOLOGICAL STUDIES

SECTION I

OBSERVATIONS ON THE BIOLOGY AND PHENOLOGY OF *PARTHENOLECANIUM RUFULUM* (HEMIPTERA: COCCIDAE) IN NORTH-EASTERN ITALY WITH A REDESCRIPTION OF THE FIRST AND SECOND-INSTAR NYMPHS

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Introduction

Parthenolecanium rufulum, the oak soft scale, is a Palaearctic species, common on *Quercus*, but also recorded on *Carpinus, Castanea, Corylus, Fagus, Sarothamnus, Robinia, Rosa, Ulmus, Vaccinum and Rubus* (Ben-Dov, 1993?). It is distributed throughout Europe, with the exception of Albania, Ireland and Portugal where it is until now unrecorded (Ben-Dov & Miller, 2008). *P. rufulum* is also unrecorded in the Northern -European countries, where deciduous oaks are at the northern border of their range.

Some other soft scales also can live on European deciduous oaks, namely *Eulecanium ciliatum* (Douglas), *E. tiliae* (Linnaeus) and, *Parthenolecanium corni* (Bouché), but. Among them *P. rufulum* is the most common and widespread species. Until a few years ago, *P. rufulum* was considered to be of no economic importance, but recently it has been reported as a pest of hazelnut (*Corylus avellana* L.) in Turkey (Saruhan & Tuncer, 2001) and as infesting deciduous oaks in urban environments in Georgia (Tbilisi) and Turkey (Ankara) (Japoshvili, 2001; Ülgentürk, 2004). Deciduous oaks are becoming of increasing their economic importance as ornamental trees in Italy where they and are currentlyat present largely used in urban parks and gardens. Despite its wide distribution, the only known biological data on *P. rufulum* relates toconcern Central Europe, namely Germany and Poland (Schmutterer, 1954, 1972; Dziedzicka, 1968). Due to this fact, observations on its phenology and biology were therefore have been carried out in North-eastern Italy.

Among the natural enemies of *P. rufulum*, *Anthribus nebulosus* Forster (Coleoptera: Anthribidae) is regarded as an effective predator in Central Europe. Its phenology is closely related to that of its prey, whichthat consists of monovoltine soft scales and their eggs. According to Kosztarab & Kozár (1983) and Ponsonby & Copland (1997), adult *A. nebulosus* appear inat mid April and feed on all stages of soft scales, plus their honeydew and fungi. The eggs are laid singly, from early May to early June, under the body of the female scale. The beetle larvae are active from mid June to July, mainly feeding on the scale eggs and pupate then under the scale cover. The newly emerged adults enter diapause in August, in sheltered places, where they remain until the following spring. Because of its potential importance as a natural enemy, observations

were also made on on the possible occurrence and the activity of *A. nebulosus* were also planned in the monitored area.

Species identification of scale insects is usually based on teneral adult female morphology. In the absence of adult females, slide mounted young instars, which may be present on the host plant for most of the year, can help in identifying the species. Among the species of *Parthenolecanium* recorded in Europe, *P. corni* is highly polyphagous and shares with *P. rufulum* several host plants, on which both the species can live together (Stepaniuk & Lagowska, 2006). Young instars of *P. corni* have been described by Schmutterer (1954), Canard (1960), Dziedzicka (1968) and Tereznikova (1981), but those ofwhereas *P. rufulum* nymphal instars were only shortlyhave only been briefly described by Schmutterer (1954). A further short description and illustration of *P. rufulum* 2nd-instar nymph was published by Dziedzicka (1968). With the collection of fresh specimens, the opportunity is here taken to redescribe and illustrate the 1st and the 2nd-instar nymphs of this species, according to up to date standards, the 1st and the 2nd-instar nymphs of this species and to discuss the differences from those of *P. corni*.

Materials and methods

This study was carried out in the park of the Faculty of Agriculture, University of Padua, North-eastern Italy, from July 2006 to July 2008. Random samples were collected on 50 twenty-year old Quercus robur trees, 20 years old, each about 7 m high. Samples were collected weekly in April, May and June and every 10-15 day during the other months. A total of about 3m of 1-2 years old branches, 1-2 years old, were sampled and all the scales, both those settled on the leaves or on the wood of the sampled branch, depending on the season, were counted. Frequencies were calculated over the total length of the sampled branches and referred to one meter and ten-days periods; the population dynamics were plotted on logarithmic scale graphs in order to showmark the abundance of each instars occurring on the branches at lower frequencies than those on leaves. Data on temperature and rainfall were provided by the local weather station (Graphs 1-3). Fecundity of 10 adult females and the egg hatching-time were checked in laboratory. Fully-grown adult females (which had

therefore stopped feeding), were collected in the field at the beginning of oviposition, and were placed on petri-dishes transferred under laboratory conditions (T° 25 \pm 0.5° C, $70 \pm 3\%$ RH, and photoperiod of 16:8 (L:D)). Eggs were counted every day until the end of oviposition. The incubation period and hatching rate were recorded. The occurrence of the predator Anthribus nebulosus was monitored between in May and-July in 2007 and 2008. With regard to the morphological description, all specimens were slide mounted according to the procedures of Ben-Dov and Hodgson (1997). Measurements and numbers are given as ranges, followed by the mean in parentheses. Terminology follows that of Hodgson (1994).

Graph 1. Climatogram of the research area in 2006.



2006

Graph 2. Climatogram of the research area in 2007.



Graph 3. Climatogram of the research area in 2008.



2008



PLATE 1. (a) First - instar nymphs of *P. rufulum* Cockerell on leaves undersurface; (b) 1^{st} instar nymph and (c) 2^{nd} instar nymphs on oak leaf during summer; (d) overwintering 2^{nd} instars on twigs; Comparison between (e) 2^{nd} instar nymph after overwintering and (f) young female (reddish); post reproductive females (g) and teneral adult females before ovipositing (h). (i) Oak twig with heavy *P. rufulum* infestation; (j) *Anthribus nebulosus* feeding on *P. rufulum* adult.

RESULTS

Observations on phenology and biology of P. rufulum

Year 2006 (Fig. 1)

Sampling started in midJuly, when the 1st-instar nymphs were already settled on the undersurface of the leaves. First instars scored a mean frequency of 2124 specimens/m of leaf Moulting to the 2nd-instar was first noted in lateAugust and, by early September, about 60% of the population were 2nd-instar nymph and by late September, the whole scale population was 2nd-instars, with a mean frequency of 602 nymphs/m From mid September, the 2nd-instar nymphs gradually moved from the leaves to the twigs to overwinter. By late September, at least 70 % of the 2nd-instars were already settled on the branches and this migration was completed by mid-December. An average frequency of 30 nymphs/m was recorded on branches during winter.



Fig. 1. Population dynamics of P.rufulum on *Quercus robur*, in the Padua district, Italy, year 2006.

After overwintering, the 2nd-instar nymphs moulted to adult females, starting in mid April, with a peak adult female abundance in mid April, when they constituted 58 % of the population. After mid May, the entire scale population was adult females, with an average frequency of 11 females/m. No males were observed, so it is confirmed that reproduction is parthenogenetic. Just after the last moult, the adult females were flat, 1.6-1.8 mm long and 0.9-1.2 mm wide, but they then matured rapidly, becaming convex, and about 6mm long and 4 mm wide. The first egg-laying females were observed in late April. In early May, egg-laying females constituted about 53 % of the population and, from mid May onwards, all adult females were egg-laying. Egghatching started in late May and ended a week later. After emergence, the crawlers moved from the twigs to the undersurface of the leaves, where they settled near the main leaf veins. First instars scored an average frequency of 1830 instars/m. Second instars were observed from August 10th, and the whole population was second instars by August 24th, when they had an average frequency of 536 instars/m. The migration from leaves to twigs started in late October and the whole population was settled on branches by mid November, with a mean frequency of 24 nymphs/m. In laboratory, an average fecundity of 1892 eggs/female was observed (minimum 425; maximum 2410 eggs/ female).



Fig. 2. Population dynamics of *P. rufulum* on *Quercus robur*, in the Padua district, Italy, year 2007.

Year 2008 (Fig. 3)

After overwintering, the 2nd-instar nymphs began moulting into adult females in mid April, as in the previous year, and adult females constituted 80 % of population by early May. Moulting was complete by mid May, with an average frequency of 8 adult females/m. The first eggs were observed in mid May and all the females were egg-laying, from late May onwards. Crawlers started to emerge in early June and hatching lasted about 10 days. The 1st-instar nymphs settled on the leaves, attaining an average frequency of 1319 nymphs/m. In the laboratory, an average fecundity of 333/eggs female was recorded (minimum 226; maximum 487 eggs/ female). Oviposition lasted on average 6.8 days; the incubation period was 6.7 ± 2.8 days, with a hatch rate close to 100 %.



Fig. 3. Population dynamics of *P.rufulum* on *Quercus robur*, in the Padua district, Italy, year 2008.

Observations on Anthribus nebulosus

Between May and July in 2007, a total of 270 egg-laying and post-reproductive *P*. *rufulum* females were observed. Of these, 106 had a larva, pupa or an adult *A*. *nebulosus* within the egg chamber, with a predation rate of 39%. Larval and pupal *A*. *nebulosus* were recorded from May 25th until mid June. Adult predators were observed on June 5th and 18th and on July 3rd. In the same period in 2008, a total of 178 female *P. rufulum* were observed but only 26 had a larva, pupa or an adult predator within the egg chamber, giving a predation rate of only 14.6%. *A. nebulosus* pupae and adults were observed from June 12th until the end of the month.

Morphology



Fig. 4. *Parthenolecanium rufulum*, 1st-instar nymph.

Where A = antenna B = spiracular trilocular disc-pore; C = metathoracic leg; D = abdominal ventral seta; E = dorsal simple pore; F = marginal seta; G = dorsal trilocular pore.

Description of 1st instar nymph (Fig. 4)

Mounted material: described from 5 specimens in good conditions, details checked on the remaining specimens; body oval; crawler body length: 370-389 (379) μ m, width: 207-215 (211) μ m; body length before moulting to 2nd instar: 673-685 (679) μ m; anal cleft short.

Dorsum: derm membranous. Dorsal simple pores, each less than 1 μ m wide, scattered along body margin except anterior 1/3 of margin where they are absent. One pair of trilocular pores, each about 2 μ m wide, situated on head apex. Anal plates each sub-triangular, broad, with inner margins slightly diverging; each plate with 1 posterior margin seta, 10-16 (13) μ m long, 2 inner margin setae, each seta 6-9 (6) μ m long, with one flagellate apical setae 174-196 (187) μ m. Ano-genital fold with 1 pair of anterior margin setae, each 10-30 (18 μ m) long, and 1 pair of lateral margin setae, each 10-13 (11) μ m long. Anal ring with 3 pairs of setae.

Margin: marginal setae setose, each 10-13 (12) μ m long, seta set in a basal socket 2-4 (3) μ m wide, distributed along body margin as follows: 6-8 anteriorly between eyespots; 2 between eyespot and anterior stigmatic area; 2 between stigmatic areas; 8 between posterior stigmatic area and anal cleft. Stigmatic spines: 3 per cleft, each slightly shorter and more spinose than marginal setae, set slightly onto dorsum, with median spine longer than laterals.

Venter: derm membranous, segmentation obscure. Ventral setae, each 3-6 (4 μ m) long, present in submarginal and submedial rows; setae mostly present on last abdominal segments; also with one pair of inter-antennal setae, each 17-26 (22) μ m long; with 1 pair of pregenital setae, each 23-25 (24) μ m long. Antennae 6-segmented, each 122-137 (129) μ m long, third segment longest. Spiracles: peritremes 5-7 μ m wide. With 3

spiracular disc-pores, each usually with 3 loculi, 3-5 (4) μ m wide, forming a short band from each spiracle to body margin. Legs well developed, without a tibio-tarsal sclerosis; claw denticle small; with setose claw digitules tapering at their base, 1.6-2 (1.8) μ m long; tarsal digitules each 23-36 (31) μ m long.

Description of 2^{nd} instar nymph (Fig. 5)

Mounted material: described from 11 specimens in good conditions, details checked on the remaining specimens; body elongate oval; dimensions after the first moult: body length: 760-969 (837 μ m), body width: 447-566 (501) μ m; dimensions before moulting to adult female: body length: 1669-1788 (1746) μ m, body width: 938-1147 (995) μ m; anal cleft short.

Dorsum: derm membranous. Dorsal microducts minute, present throughout, each 1.5 μ m wide, with a filamentous inner ductule and sclerotised pore, appearing bilocular when viewed from above. Anal plates each subtriangular, broad, with inner margins slightly diverging; anal plate setae as follows: 1 posterior margin seta, 9-13 (12) μ m long, 2 inner margin setae, latter in a subapical position, each 7-12 (9) μ m long, 1 apical seta, 9-15 (12) μ m long. Anogenital fold with 2 pairs of anterior margin setae, 12-15 (13) μ m long and 16-22 (20) μ m long respectively, and 1 pair of lateral margin setae 15 μ m long. Anal ring with 3 pairs of setae.

Margin: marginal setae sharply spinose, each 10-13 (12) μ m long, 1.5-2 (1.7) μ m wide at base, each seta set in a basal-socket 4-6 (5) μ m wide, distributed along body margin as follows: 13-14 anteriorly between eyespots; 5-7 between eyespot and anterior stigmatic area; 6-7 between stigmatic areas; 16 between posterior stigmatic area and anal cleft. Stigmatic spines: 3 per cleft, each more spinose and longer than marginal setae, median spine bluntly spinose, 22-25 (23) μ m long and 3-4 μ m wide at base, set slightly onto dorsum, lateral setae shorter, each 12-17(14) μ m long and 2-4 μ m wide at base. With 1 seta on each anal lobe significantly longer than other marginal setae, each 23-35 (29) μ m long. Width of each eyespot lens 15-16 μ m.

Venter: derm membranous, segmentation obvious medially on abdomen, obscure elsewhere; with minute dermal spinules appearing most frequent medially around anal cleft. Short ventral setae, each 4-9 (6) µm long, present in submarginal and submedial rows along body margin, mainly distributed on abdominal segments; also with 3 pairs of pregenital setae 42-58 (50) µm long; plus two pairs of inter-antennal setae: one pair 22-33 (27) µm long and the other 104-118 (113) µm long. Ventral microducts forming a band 2-3 pores wide along body margins, each microduct with a sclerotised pore 1.5-3 (2) µm wide, with short and broad inner ductule. Antennae 6-segmented, each 170-204 (191) µm long, 3rd segment longest, usually with 1 distinct pseudo-segmentation, causing antenna to appear 7-segmented; apical segment with 3 or 4 antennal bristles, 3 or 4 fleshy setae and 3 or 4 flagellate setae, longest 61-62 µm long. Spiracles: peritremes 12-15 (13) µm wide. Spiracular disc-pores quinquelocular pores, each 3-4 µm wide, forming a band of 9-12 (10) pores between each spiracle to body margin. Other ventral pores: with one pair of preantennal simple pores, each about 3 µm wide, near each scape. Legs well developed, without tibio-tarsal scleroses; claws denticles small; claw digitules unequal: one broad, 20-25 (23) µm long, and other setose, 22-28 (23) μm long; tarsal digitules capitate, each 33-39 (36) μm long.

Comments: The nymphs of P. corni and P. rufulum can be easily separated P. corni 1st-instars have two longitudinal lines of bilocular pores dorsally which are absent on P. rufulum 1st-instars. And second instar P. corni always have dorsal submarginal tubercles, although the number may be variable (Dziedzicka & Sermak, 1967); these are always absent on second instars P. rufulum. The same nymphal instars of. E. ciliatum and E. tiliae, that may occur on the same host plants as P. rufulum, have not yet been described.

Material examined: ITALY: Padua, *Quercus robur*, 24. VII. 2007: 11 1st-instar nymphs; Padua, *Quercus robur*, 24. VIII. 2006: 6 2nd instar nymphs; Padua, *Quercus robur*, 22. III. 2007: 29 2nd-instar nymphs.

Fig. 5. *Parthenolecanium rufulum*, 2nd-instar nymph.



Where A = antenna; B = ventral view of anterior stigmatic area with quinquelocular disc-pore and stigmatic setae; C = marginal band dorsal microducts; D = ventral dermal spinules; E = abdominal ventral seta; F = dorsal view of anal plates; G =. dorsal microduct.

Discussion

This survey confirms that P. rufulum is a parthenogenetic species that has one generation/year and overwinters as 2nd-instar nymphs. These 2nd-instars move from the leaves towards the branches in the Autumn, where they settle. Once settled, they do not move for the remaining part of their life, with the final moult to adult female and oviposition taking place where the nymphs settled in the past Autumn. There is no spring migration of 2nd-instars from the overwintering sites as happens with P. corni (Canard, 1958).

As was predictable, the phenology pattern of P. rufulum in Northern Italy appears earlier than that reported for Central Europe by Schmutterer (1954, 1972) and Dziedzicka (1968). In Italy, egg-laying started in late April in 2007 and in mid May in 2008, rather than between the end of May and the end of June, as reported for Central Europe. In Italy, the eggs hatched in late May in 2007 and in early June in 2008, rather than between the end of June, as in Central Europe. The delay in oviposition and eggs hatching observed in 2008 with respect to 2007 is probably due to the unfavourable weather conditions during the Spring of 2008, as reported below.

The mean fecundity was noticeably higher in 2007 (average 1892 eggs/female), than in 2008 (average 333 eggs/female). The lower fecundity observed in 2008 may be due, in part, to the smaller body size attained by adult females in comparison with 2007. In 2007, the preovipositing females reached about 6 mm in length and 4 mm in width, while in 2008, they were on average 4.3 mm long and 2.7 wide. It is hypothesized that the smaller size and subsequent lower fecundity observed in 2008 were affected by the unfavourable spring weather conditions during the growing period of females. The average monthly minimum of temperature in April and May 2007 was 9.6°C and 13.8°C, whereas it was 7.7°C and 12°C for the same months in 2008; moreover, the average maximum of temperature in April and May 2007 was higher, 22.3°C and 24.5°C, whereas it was17°C and 22.6°C) for the same months in 2008. With regard to rainfall, April and May 2007 were sunny, with only 1 and 6 rainy days/month respectively, whereas these months were colder, unusually overcast and rainy in 2008, with 13 and 22 rainy days respectively.

The data for fecundity differs considerably not only between years but also with regard to that observed by Schmutterer (1972) in Germany (about 800 eggs/female) and by Dziedzicka, (1968) in Poland (about 700 eggs/female).

According to our the data, the population levels appears rather stable, with an average frequency of 11 females/m in 2007 and 8 females/m in 2008. A high mortality was observed during the first instar, but this is quite normal. A further high mortality occurred during the migration of 2nd-instar nymphs from the leaves to the twigs..

Data on predation by A. nebulosus differed in the two years: in 2008, the predation rate was about 40%, whereas it reached only 14.6 in 2008. However, a two year observation period is too short to assess the incidence of this predator on the scale population.

Further observations are needed to obtain reliable data on female fecundity in North Italy and on the impact of predation. **SECTION II**

BIO-ECOLOGICAL STUDIES ON PSEUDOCOCCUS COMSTOCKI (KUWANA) (HEMIPTERA: PSEUDOCOCCIDAE)

Introduction

Pseudococcus comstocki was first described by Kuwana (1902) and recently redescribed by Williams and Granara de Willink (1992). It was recorded in Japan on mulberry and is currently common in China, Korea and Japan; from where it was incidentally introduced to other countries. Hence the origin of *P. comstocki* is the Oriental Region (Blumberg, 1999; Kostztarab, 1996; Meyerdirk and Newell, 1979). In the past *P. comstocki* has been confused with other mealybugs, particularly with *Pseudococcus cryptus* Hempel (Pellizzari, 2005; Blumberg, 1999; Heidari, 1999).

P. comstocki is a highly polyphagous scale insect and a widespread pest of fruit trees and ornamentals (Heidari, 1999; Kostztarab, 1996). It is currently recorded on 42 different botanical families (Ben Dov, 2008). It is a notorious pest of ornamentals (e.g. mulberry and umbrella catalpa) and fruit trees (e.g. apple, pear, peach, citrus orchards) in the Countries where this oriental species has been incidentally introduced (e.g. USA, Canada and Argentina) (Blumberg, 1999; Kostztarab, 1996; Shoene, 1939; Hough, 1925). In the Far East Countries, that are believed to represent its native area, P. comstocki is regarded as a pest of commercial fruit crops and heavy infestations are reported on high-income "bagged" apple cultivations (Bartlett, 1976). Damage is due to trophic activity and by the excretion of large quantities of honeydew, with resultant growth of sooty mould fungi. These moulds reduce photosynthesis and result in a downgrading of fruits, and are therefore an additional cause of economic loss. The production of waxy material is detrimental to ornamentals; moreover the mealybug feeding activity by injection of toxic saliva causes on mulberry the formation of galllike malformations on twigs (Kostztarab, 1996). In the orchards most of concerning is due to the fruit infestation at harvest, when the adult females are often concealed in the peduncolar or calyx end of fruits. After picking, the adults, or eventually crawlers, emerge from their shelter and their occurrence cause quality downgrading. Fruit infestations at harvest may be also noxious to fruits processed to be made into puree, because unacceptable insect contaminants may be found in the finished product (Agnello et al., 1992). Finally, this way the mealybugs can easily be transported far away with fruit trade.

This species was at first recorded in central-western Europe (Italy) in 2004 on mulberry trees (*Morus nigra*) in the district of Verona (Veneto Region) (Pellizzari, 2005). In the same year it was also collected in France on *Morus kagayamae* in urban environment (Kreiter & Germain, 2005). Subsequent records were from Treviso, on ornamentals, and Verona (Veneto Region) in 2006 and 2007, when a severe infestation was detected in a peach orchard (Pellizari *et al.*, 2007).

Because of the economic importance of *P. comstocki*, many papers have dealt with its life history in the USA, where the pest has been introduced (Agnello, 1992; Meyerdirk and Newell, 1979; Hough, 1925). Furthermore, demographic parameters and adaptations to different temperature regimes were studied in laboratory by Heidari (1999).

Due to the fact that infestation foci in the Verona district currently represent a threat to peach growers, a comprehensive study on the bio-ecology of *P. comstocki* has been carried out. A preliminary survey on the occurrence and distribution of the Comstock mealybug was assessed in the Verona district (subsection I). Studies on parasitoids and on the effect of different treatments on *P. comstocki* were also planned (subsection II and IV), in order to suggest guidelines for pest management. In addition to fieldwork, several life history and demographical parameters were observed on *P. comstocki* colonies reared under screen-house and laboratory conditions, at two temperature levels (subsection III).

Materials and methods: Study site

The present study was carried out in the Verona district, North eastern Italy, where most samplings were collected off mulberry trees, peach orchards and ornamentals (fig. 1). Further samplings were carried out in Treviso on ornamentals in urban environment. The Verona district lies in the Po valley, north-eastern Italy, in an highly urbanised area characterised by peach and nectarine cultivations that currently represent an important income sources. Most of the observations were carried out on a orchard located in the "Alpo" area, Villafranca di Verona, Verona district, described in subsection I. This site has been chosen since a heavy infestation of *P. comstocki* was recorded in August 2007, but, according to the grower, the first outbreak was noticed in 2006.



Fig. 1. Maps showing the study site in Verona, Veneto region, North-eastern Italy. The infested peach orchard located in "Alpo", Villafranca di Verona, is indicated by the red spot (map from http://www.maps.google.it/).

SUBSECTION I

ASSESSING SPATIAL DISTRIBUTION OF *P. COMSTOCKI* IN NORTH-EASTERN ITALY

Introduction

Since its first record in Europe on mulberry (Morus spp) in 2004 (Pellizzari, 2005; Kreiter & Germain, 2005), P. comstocki has been recorded on several ornamental plants in urban environment in Treviso and in several locations within the Verona district, but only in late 2006 it was detected in a commercial peach orchard near Villafranca di Verona (Pellizzari et al., 2007). The orchard is located in the "Alpo" area, Villafranca di Verona [N 45° 23.774 E 10° 56.620] (fig.1). and is here named "Alpo" orchard. The 2.5 ha plot was planted in 1993 with peach and nectarine cv at about 1500 plants/ ha (5.5 µm between rows and 3 µm between plants). It lies on a nearly flat plateau, has permanent natural ground cover between rows and is surrounded by other peach orchards, except at its Northern edge, where kiwifruit (Actinidia deliciosa) orchards occur, and Western edge, where the border supports a spontaneous mixed grassland beyond one kiwifruit hedge. It is managed according to traditional farming standards. Due to the nuisance of P. comstocki as a pest of fruit orchards, it was at first important to know its distribution in the Verona district. Finally this study focused on the "Alpo" orchard in order to gather data on the distribution of the pest inside the orchard.

Material and methods

Sampling area was centred in the infested peach orchard "Alpo". Sixteen 2,5 x 2,5 km sampling plots were disposed on a North-to-South oriented, 10 x 10 km regular, sampling grid (Fig. 2). The monitored area was eventually widened by the addition of further 8 2,5 x 2,5 km plots at the south side of the previous square sampling area. Samplings were performed weekly during summer, starting from 10.06.2008 to

28.08.2008. Each plot was sampled at least once. Monitoring focused on mulberry trees (*Morus nigra*), since they are widely distributed in the Veneto Region and are known as preferred host plant of *P. comstocki*. Additional samplings, based upon previous unverified records, were carried out in Verona district (Castelnuovo del Garda, Lugagnano, S. Martino Buon Albergo, S. Giovanni Lupatoto) and Treviso. Species identification was based on slide-mounted specimens.

In addition, the distribution of infested trees within the "Alpo" peach orchard, was assessed during summer 2008 by spatial distribution analysis. Samplings were performed from 10.07.2008 to 17.07.2008. The occurrence of *P. comstocki* was monitored by qualitative visual check of each plant and *P. comstocki* abundance is here computed after three infestation classes: 0 for "no infestation or plant absent", 1 for "infested plant" and 2 for "highly infested plant". The resulting spatial distribution is here represented by bubble plot. Data spatial autocorrelation were explored and Moran's spatial index of aggregation (Moran's I) computed with *Rookcase* (Inouye, 1999) using a randomisation statistical test.

Results

Distribution in the Verona District (Fig. 2)

P. comstocki was recorded in almost all the sampled areas in the Verona district. Infestations on peach orchards were recorded in two other sites: Lugagnano and Dieci Bine (Sona). The other sites are Cà di David, Marchesino Bovo, Verona, Alpo di Dossobuono, Villafranca di Verona, Rizza, Castel d'Azzano, Buttapietra. Most of samples were collected off mulberry (*Morus nigra*) Further samples were collected off ornamentals (i.e. *Viburnum tinus* and *Prunus laurocerasus*). In addition to the foci occurring within the sampling grid, further records regard San Martino Buon Albergo on *Viburum tinus*, San Giovanni Lupatoto on *Morus nigra*, Castenuovo del Garda on *Hypericum* spp.. These data gather with previous records from Treviso: in summer 2006 infestations were discovered on several ornamentals (*Carpinus betulus*, *Cotoneaster horizontalis, Hedera helix, Mahonia aquifolium, Pittosporum tobira, Prunus laurocerasus, Pyracantha coccinea, Viburnum tinus, Eleagnus spp.*) in urban environment. **Fig. 2.** Sampled area in the Verona district (North-Eastern Italy). Infestation foci are represented by red scores, null samplings are indicated in blue.



High infestations were observed on *P. laurocerasus* and *Viburnum tinus* hedges, where leaf-drop was observed. Furthermore, specimens of mealybugs collected on apple (*Malus domestica*) in Modena and Bologna districts (Emilia Romagna Region) during 2006 and 2007, and sent for identification to the Department of Environmental Agronomy and Crop Science, University of Padua, proved to be *P. comstocki*, suggesting a wide distribution of the pest throughout North Italy.

Infestation distribution within study site "Alpo" (Fig. 3)

A map of distribution *P. comstocki* in the "Alpo" orchard is provided. Most of the highly infested trees occurred at the centre of the orchard, whereas the trees along borders are almost uninfested. Positive correlation in spatial distribution of each score was observed (Moran's I = 0.647; lag distance = 3m).

Fig. 3. Distribution of severe infestation within peach orchard in "Alpo" area, Villafranca di Verona, Verona. Dimensions of each record (sampled tree) follow infestation level, as indicated in the legend.



Discussion

As it was predictable, in few years *P. comstocki* has become a widespread species. Foci occurred in most of the sampled plots and in many locations both in the Veneto Region (Verona and Treviso districts) and in the Emilia Romagna Region (Modena and Bologna districts). For these reasons monitoring and control measures are recommended at landscape level before to experience more severe outbreaks and agricultural economic losses. Pest management strategies should take into account the high incidence of *P. comstocki* on mulberry trees (*Morus nigra*); this implies that non-crop habitats may sustain mealybug populations and are infestation sources for nearby fruit orchards.

Field margins do not represent only a source of infestation. Several studies on the border effect focused on the influence of the surrounding vegetation (hedges, woodlands and grasslands) as a source of natural enemies (Altieri *et al.*, 2003). With regard to the present study, the consistent spatial distribution of infestation in the Alpo orchard could be ascribed to the activity of natural enemies originating from the untreated field margins of the orchard. More experiments should be conducted to confirm the spatial distribution of *P. cosmtocki* within the orchard, together with a survey on the ecological factors that might affect it.

SUBSECTION II

BIOLOGY OF *P. COMSTOCKI* UNDER SCREEN-HOUSE AND LABORATORIAL CONDITIONS

Introduction

Due to the economic importance of *P. comstocki* as a pest of fruit orchards and ornamental plants, several bio-ecological studies were carried out in the Countries where this polyphagous pest has been introduced. Most of the studies on *P. comstocki* life history were conducted in the USA: in California *P. comstocki* develops three (Bartlett, 1972) or four (Meyerdirk & Newell, 1979) generations per year. Other studies, carried out in Virginia on apple (Schoene, 1939) and Catalpa (Hough, 1925), report three generatios per year. In addition, the effect of temperature on demographic parameters was investigated by Heidari (1999) under laboratory conditions.

No data are available for both Italian or European environments. Because there is a high possibility of *P. comstocki* spreading to areas around current infestation foci (subsection I), the knowledge of its phenology and life history is of strategic importance. Such information play an important role in pest management in order to apply chemical and biological control methods.

In this study the phenology of *P. comstocki* has been outlined on potted plants culitivated under screen-house, moreover life table parameters have been obtained in laboratory at two temperature regimes.

Biology of P. comstocki under screen-house conditions

Materials and methods

Captive populations of *P comstocki* were established in order to assess mealybug phenology. Fieldwork was carried out at University of Padua, Faculty of Agriculture, North-eastern Italy, from September 2006 to December 2008. The rearing started from overwintering eggs collected outdoors, off infested *Prunus persica* and *P. laurocerasus*. Insects were reared on potted *Prunus persica*, *Malus domestica*, *Pyrus*

communis and *Morus nigra* trees in a screen-house measuring 7,10 m x 2 m x 3,40 m. Pots were isolated from the floor in order to avoid ants. Observations were carried out weekly during spring and summer and every 10-15 days during the other months. *P. comstocki* developed on all four species of host plants; no significant differences emerged in the life history parameters and data were subsequently regarded as means of all the host plants. During 2008 it was necessary to re-introduce females or eggsacs, due to the activity of hymenoperan parasitoids that caused a drop in the population level. Life-history pattern of 2^{nd} and 3^{rd} instar nymphs were assessed together due to the difficulty to separate these instars by visual observation on the tree. In fact their identification is possible only on slide-mounted specimens. Fecundity was assessed with daily observations on batches of 10 adult females collected outdoors off *P. persica* and *Hypericum* spp on October 2007 and 2008 and kept under screen-house conditions. Further data on adult females. Shelters for ovipositing females were provided by sheets of waved cardboard (15 x 30 cm) wrapped around the trunks.

Results

Phenology (Graph. 1)

The results of two years of observations are reported in graph. 1, where the phenology patterns during 2007 and 2008 are provided. According to the present survey, *P. comstocki* develops three generation/year and overwinters at the egg stage. The eggs are laid in masses with waxy filamentous secretions in protected places, such as under bark crevices or near pruning cuts. After overwintering, egg hatching started from early April (April 9th) in 2007 and from the end of April (April 28th) in 2008. During the first week after hatch 1st-instar nymphs (crawlers) infest leaves and flower buds. Moult to 2nd instar nymphs occurred during early May (May 9th) in 2007 and mid May (May 17th) in 2008. The whole population completed moulting to 2nd-instars in May 17th 2007 and June 5th 2008. The moults to adult females started during the first days of June, namely June 1st in 2007 and June 12th in 2008. In early June (June 5th, 2007)

and end of June (June 24th, 2008) all the scale population was made up of adult females. Egg-laying was observed between mid June (June 20th) in 2007 and early July (July 2nd) in 2008. Egg hatching started from the end of June (June 24th) in 2007 and mid July (July 15th) in 2008. In 2007 and 2008 adult females of the 2nd generation were observed from the beginning of August onward (August 6th 2007 and August 5th 2008) and their occurrence covered most of September. Egg laying activity, that originates the 3rd generation, occurred from August 13th until September 18th in 2007 and from August 11th until September 9th in 2008. With regard to the last generation, moult to adult females occurred in October (October 15th in 2007 and October 8th in 2008). Oviposition began in October, namely October 22th in 2007 and October 15th in 2008.

Graph. 1. Phenology of *Pseudococcus comstocki* (Kuwana) under screen-house conditions in the Padua district, Italy, years 2007 and 2008.



Fecundity (Graph. 2)

Oviposition period lasts 3-13 (7± 4.46) day. A mean fecundity of 78- 102 (89 ± 12) eggs/ female was scored and no significant difference (a< 0.05) was observed among the host-plants in the two years (graph. 2). A maximum daily fecundity of 32-55 (43 ± 11) eggs/female/day was scored and > 90 % of total laid eggs is scored after 4 days. Adult female longevity is of 10-14 (13.33 ± 1.22) days (n=390): after 14 days all the females (~100%) are dead.

Graph. 2. Egg-laying activity of *Pseudococcus comstocki* (Kuwana) on *P. persica* (2007-2008) and *Hypericum* spp. (2007) (% of total laid eggs).



Discussion

In Italy *P. comstocki* develops three generations per year. It is confirmed that *it* overwinters at the egg stage, but rare adult female and immatures can be recorded also during winter months, sheltered in the bark crevices. This is confirmed by observations from previous studies in California (Meyerdirk & Newell, 1979). First instar nymphs (crawlers) of the three generations were observed in April (1st generation) between June and July (2nd generation) and in September (3rd generation), whereas the adult females occur in June, at August and October.

Chemical control should be directed against 1st-instar nymphs of the 1st-generation. They produce direct injuries to the production by infesting flower buds; furthermore, probably due to favourable temperature conditions (Heidari, 1999), mortality is lower, with respect to summer months, as discussed in the second part of subsection II. Finally, they feed on leaves, where they are exposed to chemicals (subsection III) and a higher efficacy of treatments is expected.

The knowledge of the 2^{nd} generation adult females habits is important for the pest management: adult females migrate from leaves to branches, trunk crevices and also fruits. At fruit picking many females occur on peduncolar or calyx end of fruits, from where they can move to fruits crates. This way they are easily transported away with trading. The spread of this pest at a lower scale is affected by symbiosis with ants. Ants were observed attending immatures and adult females of *P. comstocki*, both outdoors and in screenhouse. The mealybug are usually transported in the crown zone of the trunk and protective nests of soil are contructed all around.

Biology of P. comstocki under laboratory conditions

Material and methods

Some parameters of the life hystory of *P. comstocki* were studied in climatic chamber at two constant temperatures (T° of 25 ± 0.5 °C and 30 ± 0.5 °C; $70 \pm 3\%$ RH; photoperiod of 16:8 (L:D)). Mealybugs were reared on sprouted potatoes (*Solanum tuberosum*) starting from eggsacs of *P. comstocki* collected in the field (Alpo orchard). Infested potatoes were kept separately in 1.5-liter closed plastic boxes, with ventilation through holes on the cover. Each box was isolated from the others to avoid migration. At the temperature of 30 °C it was necessary to substitute periodically the wilting potatoes with new sprouted ones in order to allow mealybugs development. The eggs laid by the parental generation were followed throughout their complete developmental cycle to assess sex-ratio and survival of progeny. Mean parental fecundity was assessed and treated as sibling population density at starting point. Samples and observations were scored weekly, whereas fecundity was assessed by daily observations of 10 females. Second and 3rd instar nymphs are virtually indistinguishable and immature identification needed to be checked on slide-mounted specimens. Due to the occurrence of dense colonies, with immature stages overlapping, 2^{nd} and 3^{rd} instars dynamics and life table parameters were scored together. Developmental time and survival of 2^{nd} -instar nymphs were not separated by sex. The sex was determined starting from prepupa instars, when male instars can be easily recognised by the production of protective cocoon (male test), that begins forming at the end of the 2^{nd} instar.

Statistical analysis

Estimation of life table parameters [reproductive rate (R_0); mean generation time (T); intrinsic rate of increase (R_m); mean generation time (D); finite rate of increase (λ)] follows SAS-based procedures (SAS institute, 1999) defined by Maia *et al* (2000) and based on Jackknife methods for estimating variances and confidence intervals. The effects of two different temperatures were analysed with a two-tailed t-test (a <0.01).

Results

Temperature has significant differential effect (a < 0.01) on finite rate of increase (λ), intrinsic rate of increase (R_m) and reproductive rate (R_0 .). These life table parameters. are significantly higher on mealybugs treated with T = 25 °C, with respect to T = 30°C. The effect of temperature on *Dt* and *T* is instead low (Table 1). Furthermore, temperatures proved to have significant effect (a < 0.01) on sex ratio and reproduction of *P. comstocki*. A marked increase in sex ratio (ratio of males) was observed: it was 0.53 at T = 25 °C and 0.74 at 30 °C. Fecundity is significantly higher at T = 25 °C [68 ÷ 321 (214) eggs/ female; n =10] than at T = 30 °C [3 ÷ 69 (29,5) eggs/ female; n =10]. Furthermore, temperature affected oviposition period as well: at T = 30 °C oviposition period is 5.6 d long in average and > 90% of total eggs are laid in the first 3 days. At T = 25 °C egg-laying activity covers a two weeks period and >90 % of total laid eggs is scored after 13 days (graph. 2).

Treatment					
T = 25 °C			$T = 30 \ ^{\circ}C$		
$0.859 \pm$	0.05	b	1.818	±	0.58 a
2.230 ±	0.08	a	1.419	±	0.11 b
0.803 ±	0.04	a	0.353	±	0.08 b
147.618 \pm	42.51	a	11.033	±	4.98 b
6.420 ±	0.37	b	7.099	±	0.30 a
	$\begin{array}{c} T = \\ 0.859 \ \pm \\ 2.230 \ \pm \\ 0.803 \ \pm \\ 147.618 \ \pm \\ 6.420 \ \pm \end{array}$	$T = 25 \text{ °C}$ 0.859 ± 0.05 2.230 ± 0.08 0.803 ± 0.04 147.618 ± 42.51 6.420 ± 0.37	Treat: T = 25 °C 0.859 ± 0.05 b 2.230 ± 0.08 a 0.803 ± 0.04 a 147.618 ± 42.51 a 6.420 ± 0.37 b	Treatment $T = 25 \ ^{\circ}C$ $0.859 \pm 0.05 \ b$ 1.818 $2.230 \pm 0.08 \ a$ 1.419 $0.803 \pm 0.04 \ a$ $0.803 \pm 42.51 \ a$ $147.618 \pm 42.51 \ a$ 11.033 $6.420 \pm 0.37 \ b$ 7.099	Treatment $T = 25 \ ^{\circ}C$ T = $0.859 \pm 0.05 \ ^{\circ}b$ $1.818 \pm 2.230 \pm 0.08 \ ^{\circ}a$ $1.419 \pm 0.803 \pm 0.04 \ ^{\circ}a$ $0.803 \pm 0.04 \ ^{\circ}a$ $0.353 \pm 147.618 \pm 42.51 \ ^{\circ}a$ $11.033 \pm 6.420 \pm 0.37 \ ^{\circ}b$

Table 1. Life-table parameters (\pm SE) of *Pseudococcus comstocki* (Kuwana) reared at two different temperatures (T° of 25 ± 0.5°C and 30 ± 0.5 °C).

Graph. 3. Egg-laying activity (% of total laid eggs) of *Pseudococcus comstocki* (Kuwana) under two temperature regimes (T° of 25 ± 0.5 °C and 30 ± 0.5 °C).



Discussion

Temperature regimes affect the demographic parameters of *P. comstocki*, as reported in table 1. A constant temperature of 25 ° C was more favourable, with a net reproductive rate (R_0) of 147 females/ female, an intrinsic rate of increase (r_m) of 0.803 females/ female/ week, and a finite rate of increase (λ) of 2.230 females/ female/ week. At 30° C net reproductive rate (R_0) was 11 female/female; intrinsic rate of increase (r_m) was of 0,353 female/female/week and finite rate of increase (λ) was 1.419 females/ female/ week.

Doubling time (*Dt*) and mean generation time (*T*) were similar between treatments. According to Heidari (1999), the detrimental effect of highest temperatures (T = 30 °C) upon population growth potential, that is calculated after r_m (Maia *et al*, 2000), explains why, in field conditions, infestations are more severe in the first generation, during sprintime. Temperature was also found to have significant influence on the fecundity of *P. comstocki*. More eggs/female were produced at 25°C, moreover during a longer oviposition period, whereas, at 30°C, the oviposition rate and oviposition period were reduced. Furthermore, at 30° C a male-biased sex ratio occurred,

Although an attempt to compare the above results with bibliographical data (Heidari, 1999) is here provided and discussed, it should be minded that comparisons among life table parameters estimates cannot be afforded when differences in experimental plan or computing protocols occur. For such reasons the following discussion should be only considered descriptive and unexplanatory.

Our data seems to confirm biological data provided by Heidari (1999) in his study, even if they were obtained with different methodologies. In his study reproduction and demographic parameters are higher at 22-26 °C and lower at 30 ° C. Life table parameters obtained by Heidari (at T = 26 °C and T = 30 °C) are here reported in table 2 and compared with the present study results. A descriptive comparison between demographic parameters is allowed after a rough conversion of life table parameters, that in the present work are referred to weeks. The intrinsic rate of increase (r_m), obtained in the present study is close to that reported by Heidari at both temperatures. Significant differences were instead found among other parameters, firstly the finite
rate of increase (λ) and net reproductive rate (R_0), but they may be strongly biased by differences in experimental plan or computing protocols

Table 2. Life-table parameters of *Pseudococcus comstocki* (Kuwana) after the currentstudy (modified) and after Heidari (from Heidari, 1999).

	Treatment					
	currer	<i>it study</i>	from Heidari			
	T = 25 °C	T = 30 °C	T = 26 °C	T = 30 °C		
Dt (day) λ (female/female/day) r_m (female/female/day) Ro (female/female) T (day)	6.013 0,318 0.115 147.6 44.94	12.726 0.203 0.050 11.0 49.01	6.80 1.107 0,102 47.9 38.0	9.60 1,419 0.077 10.2 30.4		

SUBSECTION III

PARASITOIDS OF *PSEUDOCOCCUS COMSTOCKI*: FIRST RECORDS OF *CLAUSENIA PURPUREA* ISHII AND *CHRYSOPLATYCERUS SPLENDENS* (HOWARD) (HYMENOPTERA: ENCYRTIDAE) IN EUROPE

Introduction

Soon after the first oubreaks of *Pseudococcus comstocki* in the USA, where it was recorded since 1918 (Hough, 1925), several efforts were made towards biological control of the mealybug and exotic parasitoids were imported from Japan since 1939 (Schoene, 1939; Bartlett & Clacy, 1972; Meyerdirk & Newell, 1979).

The Comstock mealybug has been recorded as a pest of peach in North Italy in 2006 (Pellizzari *et al.*, 2007). Preliminary trials of chemical control (here described in subsection IV) obtained unsatisfactory results. Aiming to provide guidelines for biological control strategies in Italian agroecosystems, a preliminary survey on natural enemies of *P. comstocki* in Italy has been carried out in 2007-2008.

Materials and methods

Samplings were carried out in summer and autumn 2007-2008. Specimens of *P. comstocki* were collected off infested *P. persica* and ornamentals (*Hypericum* sp., *Morus nigra, Prunus Laurocerasus, Viburnus tinus*) in different locations in the Verona and Treviso districts. In June 2008 additional parasitoids were obtained from *P comstocki* adults reared under screen-house conditions at the Department of Environemental Agronomy and Crop Production (DEAE), as reported in subsection II. *P. comstocki* adult females, with symptoms of parasitization, were selected and transferred in glass tubes in batches of 10 specimens and kept under laboratory conditions (T° 25 \pm 0,5°C, 70 \pm 3% RH, and photoperiod of 16:8 (L:D)). Newly emerged parasitoids were collected daily. The parasitoids were identified by the specialist Dr. Emilio Guerrieri (Plant Protection Institute-CNR, Portici, Napoli, Italy).

Results

A list of the parasitoids is provided in table 1. Of them, seven species belong to the family Encyritidae and one (*Thysanus* spp.) to the family Signiforidae (Hymenoptera). The encyrtid wasp *Clausenia purpurea* Ishii was the most frequent species. Several specimens of *Clausenia purpurea*, both males and females, developed from *P. comstocki* collected off *P. persica* (Villafranca di Verona, Verona), and one *Clausenia* sp.² was obtained from *P. comstocki* collected off *Hypericum* spp. (Castelnuovo del Garda, Verona). *C. purpurea* is a new record for Europe. Moreover, the Afrotropical encyrtid wasp parasitoid *Chrysoplatycerus splendens* (Howard), swarmed from *C. comstocki* collected on *Viburnum tinus* in Treviso: it is a new record for the Palearctic Region. Furthermore, *P. comstocki* represents a new host for *Pseudaphycus maculipennis* Mercet, *Anagyrus pseudococci* (Girault), and *Anicetus africanus* (Girault, 1920).

 $^{^{2}}$ Accordino to the specialist Dr. E. Guerrieri this *Clausenia* is different from *C. purpurea* and could belong to another species.

Location Date		Host plant	Species	Family	Notes	n° records	
Verona, Villafranca	08.2007.	Prunus persica	Pseudaphycus maculipennis Mercet	Encyrtidae	Never recorded ex P.comstocki	3	
Verona, Villafranca	10.2007.	Prunus persica	<u>Clausenia purpurea Ishii</u>	Encyrtidae	new record for Europe	4	
Verona, Villafranca	10.2007	Prunus persica	Anagyrus pseudococci (Girault)	Encyrtidae	Never recorded ex P.comstocki	2	
Verona, Castelnuovo del Garda	10.2008	Hypericum spp.	Clausenia spp.	Encyrtidae		1	
Treviso	07.2007	Viburnum tinus	Pseudaphycus maculipennis Mercet	Encyrtidae	Never recorded ex P.comstocki	2	
Treviso	07.2007	Viburnum tinus	Pachyneuron sp.	Encyrtidae		I	
Treviso	9.2008	Pruus laurocerasus	Anicetus africanus (Girault, 1920)	Encyrtidae	Never recorded ex P.comstocki	1	
Treviso	10.2007	Viburnum tinus	<u>Chrysoplatycerus splendens</u> <u>(Howard)</u>	Encyrtidae	new record for the Palearctic region	1	
Treviso	10.2007	Viburnum tinus	Pseudaphycus maculipennis Mercet	Encyrtidae	Never recorded ex P.comstocki	4	
Treviso	10.2007	Viburnum tinus	Thysanus sp.	Signiforidae		I	

Table 1 . List of parasitoids ex <i>P. comstocki</i> from the Veneto Region, Italy.	mstocki from the Veneto Region, Italy.
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Discussion

Several biological control studies suggest managing *P. comstocki* outbreaks by establishment of effective natural enemies. *P. comstocki* was successfully kept under control by importation into USA of the Asiatic encyrtid *C. purpurea* as above reported. In Israel *C. purpurea* demonstrated an efficient parasitoid of mealybugs and was responsible for the control of *Pseudococcus cryptus* outbreaks during 1940' and 1990's, after its introduction from Japan (Blumberg *et al.* 1999).

During this survey (2007-2008) *C. purpurea* mostly emerged from samples of *P. comstocki,* collected from the "Alpo" orchard. Many specimens swarmed also from females bred in screen-house during July 2008, as described in subsection II. About 80% of adult females (n=325) observed on July 2^{nd} in screenhouse were parasitized and several other parasitoids swarmed in the following days. This event brought to a collapse of the colony and was necessary to start again the rearing with females collected in the field. It should be mentioned that the rearing started with eggsac with dead females collected in the Alpo infested orchard.

The pathway of arrival of *C. purpurea* in Italy is at present unknwn, but it is possible that, when introduced into Italy, *P. comstocki* was accompained by some of its main natural enemis: among the Countries of the Mediterranean basin, the intentional introduction of this species is only reported for Israel. The incidence of *C. purpurea* suggests that it is well adapted to Italian environments and, in a perspective view, it poses the basis for a future biological control of *P. comstocki*.

Chrysoplatycerus splendens is an afrotropical species, known as an effective parasitoids of mealybugs. It has been used in South Africa for successful biological control projects against *Planococcus ficus* (Summy *et al.*, 1986). *C. splendens* swarmad from *P. comstocki* collected in Treviso on ornamentals and is the first record for the Palearctic Region. Its occurrence enphasizes the possibility of biological control *P. comstocki* in Italy.

The results of this investigation demonstrate that native and exotic parasitoids are established in the infested sites. *C. purpurea* is regarded as one of the major factors that may contribute to the biological control of *P. comstocki* in Italy, when not affected by chemical treatments required for fruit pest management.

SUBSECTION IV

EFFECT OF THIAMETOXAM, CHLORPYRIPHOS-ETHIL, PHOSMET AND PYRETHRUM ON *P. COMSTOCKI*

Introduction

Infestation of *P. comstocki* occurred in a peach orchard in "Alpo", Villafranca di Verona (Verona district) (as above reported) since 2006. Most of concerning was due to the fruit infestation at harvest (August), when the adult females were often concealed in the fruit cavity at the peduncolar end, this implying fruit quality downgrading. After picking, adults can easily be transported far away with fruit trade. A trial of chemical control was performed to evaluate the effect of four insecticides on *P. comstocki*.

Materials and methods

Assays were performed in the "Alpo" orchard, near Villafranca diVerona. The orchard is managed according to traditional farming standards. The list of treatments subsequent February 2008, up to the trial (July 2008), is reported in table 1. Each chemical was used at the concentration suggested for field applications. Four active ingredients were selected for the trial because of their widespread use on fruit orchards in conventional agriculture and their expected effectiveness on scale insects. Active ingredients used during the experimental assays are listed in table 2. The treated area is a highly infested row at the centre of the peach orchard. A complete randomisation experimental set-up was established with 15 plots, each 35 m² wide. The infestation level was verified before the treatment, at 10.07.08, as frequencies of P. comstocki instars and referred to the number of examined branches; at least four branches were examined for each plant. Treatments were carried out at July 15th 2008, when 2nd- and 3rd-instar nymphs of the mealybug were the most frequent instar occurring on the trees. Occurrence of P. comstocki on treatments and control plants was checked at July 29th 2008; frequencies were calculated after the number of adult females on fruits (records) with three infestation classes (CL1 = no record; CL2 = < 3 records; CL3 = > 3 records) and referred to batches of 100 fruits for each plot.

Statistical analysis: Data were tested for normal distribution using Q-Q plot and Shapiro-Wilk test with R software program (R Development Core Team, 2008). One-way analysis of variance (ANOVA) was performed on normally distributed data using a general linear model (GLM) for all the tests.

Date	Active ingredient	Formulation	Dose [g-mL/ha]		
15.02	Ossichlorur copper	Patrol	800		
07.03	Mineral Oil	Sipcamol	2500		
	Fluvalinate	Megic	30		
	Ziram	Triscabol DG	300		
07.04.	Metomil	Lannate	200		
	Sufur	Tiosol	250		
	Ziram	Diziram	200		
07.05	Thiacloprid	Calypso	25		
	Sulfur	Tiosol	250		
	Ziram	Diziram	300		
31.05	Sulfur	Tiosol	300		
	Ziram	Diziram	250		
	Triflumuron	Retin	25		
04.07	Metomil	Lannate	250		
	Ziram	Diziram	200		
	Sulfur	Tiosol	250		

Table 1. List of treatments and active ingredients used in the examined orchard.

Table 2. List of active ingredients used during the assay.

Treatmonts	Active ingredient	Formulation	Concentration	Dose
Treatments	Active ingredient	Formulation	Concentration	[mL or g/ha]
1	Chlorpyriphos – ethil	DBursban EC	44.5%	110
2	Phosmet	Supraphos EC	17.7%	250
3	Thiametoxam	Actara WG	25.0%	30
4	Pyrethrum	Pyresan plus	4.0%	100
control	/	/	/	/

Results (table 3)

Active ingredients that shown a significant activity against the mealybug include Thiametoxan (Actara) and Chlorpyriphos-ethil (Dursban). Only the neonicotinoid Thiamehoxam increased treatment efficacy more than 50% (51,8%). Phosmet (Supraphos) and pyrethrum (Pyresan) affected poorly the occurrence of the Comstock mealybug on fruits and did not produce significant difference with respect to control.

Table 3. Effect of treatments on the occurrence of *P. comstocki* on fruits before picking. Results provided according three classes (CL1; CL1; CL3) and efficacy % (last column).

	Active ingredient	% Cl 0	% Cl 1	% Cl 2	infested fruits %	Efficacy %
1	Chlorpyriphos – ethil	48.7 ab	24.3 n.s.	27.0 bc	51.3 bc	38,2
2	Phosmet	37.3 abc	26.7	36.0 abc	62.7 abc	24,5
3	Thiametoxam	60.0 a	24.3	15.7 c	40.0 c	51,8
4	Pyrethrum	27.3 bc	31.3	41.3 ab	72.7 abc	12,4
5	Control	17.0 c	22.0	61.0 a	83.0 a	-

Different letters indicate significant differences at Tukey's Test; different capital letters indicate highly significant difference (P<0.01); Different small letters indicate a significant statistical difference (P<0.05); n.s. = not significant.

Discussion

Determining whether the mealybugs are susceptible to insecticide commonly used for pest management in peach orchards may be of strategic importance to establish a guideline for chemical control in Italian environments. Moreover treatment effectiveness requires a proper timing and the correct identification of the stages present on the host-plant is essential. because of certain mealybug habits Third-instar nymphs and adult females are known to migrate from leaves to trunk and main branches, to find shelter and lay eggs inside bark cravices. Such locations are difficult to treat with pesticides. For such reasons more satisfactory results could be obtained by chemical control against 1st-instar nymphs of the first generation. Usually 1^{st} -instar nymphs settle on leaf undersurfaces, where they are exposed to chemicals. Unfortunately, the trial was conducted in delay and treatments assessed on 2^{nd} - and 3^{rd} - instar nymphs of 2^{nd} generation, rather than on the 1^{st} -instars. The assay produced unsatisfactory results: *P. comstocki* population is not suppressed in response to the treatment, although a significant decrease in infestation rate was observed on thesis treated with Thiametoxam (Actara). Furthermore, treatments with chlorpyriphos (Dursban), that are known as effective on scale insects, led to only slight decrease in the infestation rate. Such control failure has been attributed to the above reported reasons rather than to insecticide resistance, that is currently unexplored.

CONCLUSIONS

The above reported dissertation contributes to increase the knowledge on superfamily *Coccoidea*, following both a taxonomical and ecological approach. With regard to systematics, proper morphological descriptions of several developmental stages provide new diagnostic tools for identification of four species (listed in table 1), three of which are of agricultural importance.

described species	N1	N2 female	N3	N2 male	prepupa	pupa	adult male	<i>adult</i> female
Ceroplastes japonicus				x	Х	x	x	
Ceroplastes rusci			x	X	Х	x	X	
Ceroplastodes dugesii	X	X	x	X	Х	x		X
Parthenolecainium rufulum	x	x						

Table. 1. List of descriptions and illustrations of the studied species.

Most of taxonomic work concerns the description of the adult male *Ceroplastes japonicus* Green and *Ceroplastes rusci* L. (Hemiptera: Coccidae: Ceroplastinae). The affinities with *C. ceriferus, C cirripediformis, Waxiella berliniae and Waxiella spp.* males are discussed and a first attempt of an identification key based on adult males morphology is provided. According to this morphological study, *C. rusci* and *C. ceriferus, criripediformis* with respect to *C. japonicus* and *C. ceriferus,* whereas species in *Waxiella* genus clearly are well separated from *Ceroplastes* genus.

The second part of this study provides new information on several aspects of the biology and ecology of *Parthenolecanium rufulum* (Cockerell) and *Pseudococcus comstocki* (Kuwana). These surveys may be useful to define guidelines for the pest management with both chemical and biological methods.

The study on the oak soft scale *P. rufulum* assesses the phenology of this species in Italian environments and provides new information on the bio-ecology of the scale. In addition, a descriptions of its nymph instars is provided, that could allow species identification in absence of adult females.

The study on *Pseudococcus comstocki* gives first information on the phenology and biology of this alien species in Italy. In screen-house the species demonstrates to develop three generations per year and overwinters in the egg stage. This phenology pattern was confirmed by field observations. According to the mealybug phenology, early May is regarded as the proper timing for chemical treatments, since 1st-instar nymph (crawlers) occur on the leaves. The occurrence of native and exotic encyrtid parasitoids is also reported. Among them, emphasis is given to the record of *Clausenia purpurea* Ishii, never recorded before in Europe. This Asiatic species is regarded as one of the major factors that may contribute to the biological control of *P. comstocki* in Italy. Interest also lies on the unexpected first record in the Palearctic Region of *Chrysoplatycerus splendens* Howard, known as an effective parasitoid of mealybugs in the Afrotropical Region. Records of natural enemies support the possibility of biological control of *P. comstocki* in Italy.

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